M.Sc. Final Year

Chemistry, Paper IV

CHEMISTRY OF NATURAL PRODUCTS

मध्यप्रदेश भोज (मुक्त) विश्वविद्यालय – भोपाल

MADHYA PRADESH BHOJ (OPEN) UNIVERSITY – BHOPAL

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SYLLABI-BOOK MAPPING TABLE

Chemistry of Natural Products

Syllabi	Mapping in Book
Unit - I Terpenoids and Carotenoids Classification, nomenclature, occurrence, isolation, general methods of structure determination, isoprene rule. Structure determination, stereochemistry, biosynthesis and synthesis of the following representative molecules: Citral, Geraniol, α-Terpineol, Menthol, Farnesol, Zingiberene, Santonin, Phytol, Abietic acid and β -Carotene.	**Unit-1:** Terpenoids and Carotenoids **(Pages 3-43)**
Unit - II Alkaloids Definition, nomenclature and physiological action, occurrence, isolation, general methods of structure elucidation, degradation, classification based on nitrogen heterocyclic ring, role of alkaloids in plants.	**Unit-2:** Alkaloids **(Pages 45-68)**
Unit - III Steroids Occurrence, nomenclature, basic skeleton, Diel's hydrocarbon and stereochemistry. Isolation, structure determination and synthesis of Cholesterol, Bile acids, Androsterone, Testosterone, Estrone, Progestrone, Aldosterone. Biosynthesis of steroids.	**Unit-3:** Steroid **(Pages 69-97)**
Unit - IV Plant Pigments Occurrence, nomenclature and general methods of structure determination. Isolation and synthesis of Apigenin, Luteolin, Quercentin, Myrcetin, Quercetin-3-glucoside, Vitexin, Diadzein, Butein, Aureusin, Cyanidin-7-arabinoside, Cyanidin, Hirsutidin. Biosynthesis of flavonoids: Acetate pathway and Shikimic acid pathway.	**Unit-4:** Plant Pigments **(Pages 99-125)**
Unit - V Porphyrins **Prostaglandins:** Structure and synthesis of Haemoglobin and Chlorophll. Occurrence, nomenclature, classification, biogenesis and physiological effects. Synthesis of PGE_2 and $PGF_{2\alpha}$ **Pyrethroids and Rotenones** Synthesis and reactions of Pyrethroids and Rotenones. (For structure elucidation, emphasis is to be placed on the use of spectral parameters wherever possible).	**Unit-5:** Porphyrins Prostaglandins Pyrethroids and Rotenone **(Pages 127-152)**

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UNIT 5 PORPHYRINS PROSTAGLANDINS PYRETHROIDS AND ROTENONE 127-152

INTRODUCTION

NOTES

A natural product is a chemical compound or substance produced by a living organism that is, found in nature. In the broadest sense, natural products include any substance produced by life. Natural products can also be prepared by chemical synthesis (both semisynthesis and total synthesis) and have played a central role in the development of the field of organic chemistry by providing challenging synthetic targets. The term natural product has also been extended for commercial purposes to refer to cosmetics, dietary supplements, and foods produced from natural sources without added artificial ingredients.

Within the field of organic chemistry, the definition of natural products is usually restricted to organic compounds isolated from natural sources that are produced by the pathways of primary or secondary metabolism. There are variety of natural products produced by organisms. it is important to understand the chemistry of natural products for better understanding and utilisation of these products.The terpenoids, also known as isoprenoids, are a large and diverse class of naturally occurring organic chemicals derived from the 5-carbon compound isoprene, and the isoprene polymers called terpenes. Terpenoids are the largest class of plant secondary metabolites, representing about 60% of known natural products. Carotenoids also called tetraterpenoids, are yellow, orange, and red organic pigments that are produced by plants and algae, as well as several bacteria, and fungi. Carotenoids give the characteristic color to pumpkins, carrots, parsnips, corn, tomatoes, canaries, flamingos, salmon, lobster, shrimp, and daffodils. Alkaloids are a class of basic, naturally occurring organic compounds that contain at least one nitrogen atom. This group also includes some related compounds with neutral and even weakly acidic properties.

Any of a class of natural or synthetic organic compounds which is characterized by a molecular structure of 17 carbon atoms arranged in four rings are known as steroids. Steroids are important in biology, chemistry, and medicine. Biological pigments, also known simply as pigments or bio chromes, are substances produced by living organisms that have a colour resulting from selective colour absorption. Biological pigments include plant pigments and flower pigments. A plant pigment is any type of colored substance produced by a plant. In general, any chemical compound which absorbs visible radiation between about 380 nm (violet) and 760 nm (ruby-red) is considered a pigment. There are many different plant pigments, and they are found in different classes of organic compounds. Plant pigments give color to leaves, flowers, and fruits and are also important in controlling photosynthesis, growth, and development. Porphyrins are a group of heterocyclic macrocycle organic compounds, composed of four modified pyrrole subunits interconnected at their á carbon atoms via methine bridges (=CH–). The prostaglandins are a group of physiologically active lipid compounds called eicosanoids. It has diverse hormone-like effects in animals. Prostaglandins have been found in almost every tissue in humans and other animals. They are derived enzymatically from the fatty acid arachidonic acid. A pyrethroid is an organic compound similar to the natural pyrethrins, which are produced by the flowers of pyrethrums. Rotenone is a natural compound, used as an insecticide and an herbicide.

NOTES

This book is divided into five units that attempt to give the students the basic idea of terpenoids and carotenoids, alkaloids, steroids, plant pigments, porphyrins, prostaglandins, pyrethroida and rotenones. The book follows the Self-Instructional Mode or SIM format wherein each unit begins with an 'Introduction' to the topic followed by an outline of the 'Objectives'. The detailed content is then presented in a simple and structured manner interspersed with Answers to 'Check Your Progress' questions. A list of 'Key Terms', a 'Summary' and a set of 'Self-Assessment Questions and Exercises' is also provided at the end of each unit for effective recapitulation.

UNIT 1 TERPENOIDS AND CAROTENOIDS

NOTES

Structure

1.0 INTRODUCTION

The terpenoids, also known as isoprenoids, are a large and diverse class of naturally occurring organic chemicals derived from the 5-carbon compound isoprene, and the isoprene polymers called terpenes. Terpenoids are the largest class of plant secondary metabolites, representing about 60% of known natural products. Many terpenoids have substantial pharmacological bioactivity and are therefore of interest to medicinal chemists. Terpenoids are modified terpenes, wherein methyl groups have been moved or removed, or oxygen atoms added. Just like terpenes, the terpenoids can be classified according to the number of isoprene units that comprise the parent terpene.

Carotenoids are yellow, orange, and red organic pigments. They are produced by plants and algae, as well as several bacteria, and fungi. Carotenoids are the reason for the characteristic colour of pumpkins, carrots, parsnips, corn, tomatoes, canaries, flamingos, salmon, lobster, shrimp, and daffodils. They can be produced from fats and other basic organic metabolic building blocks by all these organisms. Carotenoids from the diet are stored in the fatty tissues of animals and exclusively carnivorous animals obtain the compounds from animal fat.

NOTES

In this unit you will study about classification, nomenclature, occurrence, and isolation of terpenoids, isoprene rule, structure determination, stereochemistry, biosynthesis and synthesis of some representative molecules such as Citral, Geraniol, α-Terpeneol, Menthol, Farnesol, Zingiberene, Santonin, Phytol, Abietic Acid, carotenoids, and β Carotene.

1.1 OBJECTIVES

After going through this unit you will be able to:

- Understand classification, nomenclature, occurrence, and isolation of terpenoids,
- Interpret, the isoprene rule
- Explain the structural determination, stereochemistry, biosynthesis and synthesis of some representative molecules
- Analyse carotenoids
- Explain β Carotene

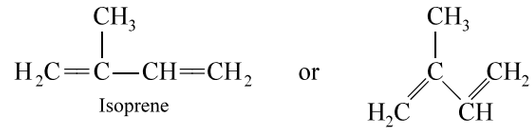
1.2 TERPENOIDS

General

Since long we are acquainted with the fragrance of flowers (*e.g.*, jasmine, rose, etc.), fruits (*e.g.*, oranges, lemons, etc.), and even wood (*e.g.*, sandal wood) of several plants especially those belonging to Coniferae and Myrtaceae families. On steam-distillation or ether extraction of various parts of these plants certain fragrant substances; finding application in perfumery, food flavouring and medicines, are obtained. These are known as *essential oils*, which may be hydrocarbons or oxygen containing substances (*e.g.*, alcohols, aldehydes, ketones, etc.). These hydrocarbons are called *terpenes*, while their oxygen derivatives are known as *camphors.* However, a more general term *terpenoid* is applied for compounds belonging to both the classes.

Structural Unit

With the exception of a few (*e.g.* totarol, eremophilone etc.), terpenes in general have the empirical formula C_5H_5 and are supposed to be derived from isoprene (C_5H_8). For this reason they are sometimes referred to as isoprenoids also.



According to Ingold (1925) molecules of terpenes are built up of isoprene units joined in a regular head to tail fashion, the head being branched end of isoprene. Thus, we see that in terpenoids the carbon skeletons of isoprene units are joined with one another through C_1 and C_4 positions. This is known as *Isoprene rule*

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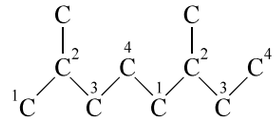
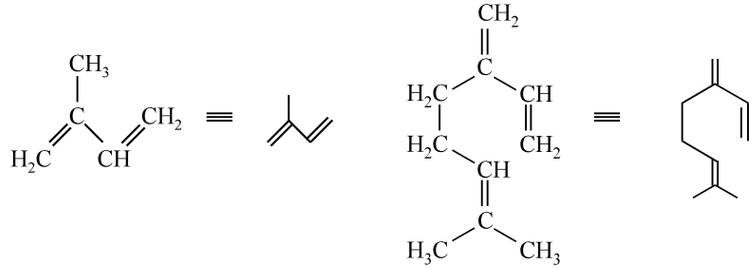
and is very helpful in the elucidation of structures of terpenoids. However, there are some terpenoids which contain carbon atoms which are not multiples of 5 and in some terpenoids head to tail joining rule is also violated.



1,4-Linkage of isoprene units

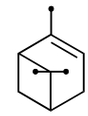
Simpler Representation of Terpenoids

For convenience sake the structures of terpenoids are represented by simpler notations. The carbon-carbon bonds are represented by line, a single line representing a single bond and two parallel lines as double bond. The carbon atoms are presumed to be at the end of these lines and hydrogen atoms are assumed to be attached to these carbon atoms in accordance with the valency requirements. For example,



Isoprene Myrcene

Sometimes a methyl group (—CH_3) is represented by a black circular dot as for example



α-Pinene

Classification

The terpenoids are classified according to the number of isoprene (C_5) units present in them. Thus we have

Hemiterpenoids	—	only one isoprene unit	C_5
Monoterpenoids	—	with two isoprene units	C_{10}
Sesquiterpenoids	—	with three isoprene units	C_{15}
Diterpenoids	—	with four isoprene units	C_{20}
Triterpenoids	—	with six isoprene units	C_{30}
Polyterpenoids	—	with several isoprene units	$(C_5)_n$

The only known hemiterpenoid is isoprene itself and that too does not occur in any botanical species.

These terpenoids are further subdivided as *acyclic* or *cyclic* depending on the fact whether they have open-chain or cyclic structures. The cyclic terpenoids may

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be *monocyclic*, *bicyclic* or *tricyclic* according to the number of rings present there in. Furthermore these terpenoids may either be hydrocarbon terpenoids or oxygenated terpenoids.

A few examples of some of the important classes are mentioned below:

(*i*) Acyclic monoterpenoid hydrocarbons, $C_{10}H_{16}$, *e.g.*, *myrcene.*

(*ii*) Acyclic polyterpenoid hydrocarbons, $(C_5H_8)_n$, *e.g. natural rubber* and *guttapercha.*

(*iii*) Monocyclic monoterpenoid hydrocarbons, $C_{10}H_{16}$, *e.g.*, *limonene.*

(*iv*) Bicyclic monoterpenoid hydrocarbons, $C_{10}H_{16}$, *e.g.*, *α-pinene*.

(*v*) Oxygenated acyclic terpenoids, *e.g.*, *geraniol, citral*, etc.

(*vi*) Oxygenated cyclic terpenoids, *e.g.*, *menthol, camphor, vitamin A* etc.

1.2.1 Isolation of Essential Oils and Terpenoids

The terpenoids occur widely in nature in different varieties of plants hence there can be no generalised procedure for their isolation. Essential oils contain many terpenoids from which the constituents can be obtained by general procedure given below. As terpenes possess a wide variety of structures the choice of the method depends on the nature of the plant, the chemical nature of individual terpene, its physical state, its sensitivity to heat and air and the use to which the essential oil is to be put.

(*i*) **Steam Distillation**. Plant material (roots, stem, leaves, flowers, etc.) is macerated and steam distilled. The steam distillate is separated, the aqueous layer saturated with salt and extracted with purified solvents, such as, light petroleum, benzene, etc. The mixture of oil and the solvent is dried and the solvent evaporated under reduced pressure to leave the oil as residue.

(*ii*) **Solvent Extraction**. The plant material is extracted directly with the solvent (ether or preferably light petroleum) at room temperature. The filtered extract is evaporated under reduced pressure when oil is left as residue.

The method is especailly useful when the oil is heat sensitive and cannot be obtained by steam distillation.

(*iii*) **Enfleurage Process**. The method is applicable for the extraction of essential oils from flowers. Flower petals are spread over an odourless mixture of tallow and lard and warmed to 50°C. They are left over for several days like this. The exhausted petals are then replaced by fresh ones. After a few weeks the fat, which has been enriched with essential oil, is freed from petals and stirred with absolute alcohol. The alcohol extract is evaporated at 0°C in vacuum to give the oil.

Unsaturated terpenes, *e.g.*, myrcene may be separated by the formation of crystalline adducts with hydrogen halides, nitrosyl chloride, etc., while the aldehydic or ketonic terpenoids may be separated as sodium bisulphite derivative or as semi-carbazide. The alcoholic terpenoids (*e.g.*, menthol) may, however, be separated by fractional distillation or by the formation of their esters with phthalic acid or as phenyl urethanes.

Terpenoids with higher molecular weight may be separated and purified by certain special methods such as chromatography, counter current method, etc.

1.2.2 General Properties of Terpenoids

1. They are generally colourless pleasant smelling liquids, lighter than water and possessing high refractive indices. A few solid terpenoids are also known, *e.g.*, camphor.

2. They are steam volatile and generally optically active.

3. They are generally insoluble in water but soluble in organic solvents.

4. Chemically they are reactive compounds and exhibit reactions due to double bond and functional groups present in them.

5. A number of them possess antiseptic properties.

1.2.3 General Procedure for Determining Structure of Terpenoids

After obtaining terpenoids in a pure state the following general procedure is adopted for determining their structure.

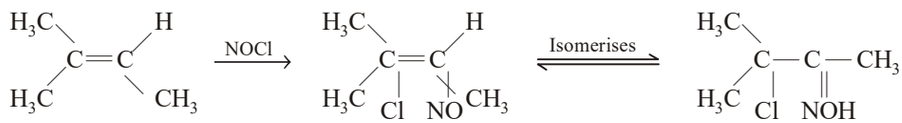
1. **Molecular formula** of the compound is determined by usual methods. This can be verified by determining molecular weight and molecular formula by mass spectrometry.

2. **Physical constants** like boiling point and melting point and solubility are determined. If the compound is optically active its specific rotation is also measured. *Optical rotatory dispersion* measurements have been found to be useful in identification and location of carbonyl group and determination of the absolute configuration.

3. **Presence of olefinic bonds** is determined by Baeyer's reagent or bromine water and their number is determined by quantitative hydrogenation, analysis of bromides, ozonolysis etc. Addition of nitrosyl chloride to compounds whose carbon atoms forming the double bond are tertiary, gives an unstable blue compound.

Blue

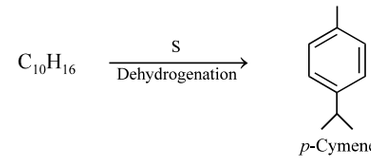
If however, one of the two carbon atoms forming the double bond is secondary the addition compounds is colourless.



(Addition according to Markovnikov's rule)

Oxime (colourless)

4. **Dehydrogenation** of terpenoids using zinc, sulphur, selenium, platinum or palladium etc. converts them into compounds of known structure. This is useful in judging the carbon skeleton of the terpenoid.



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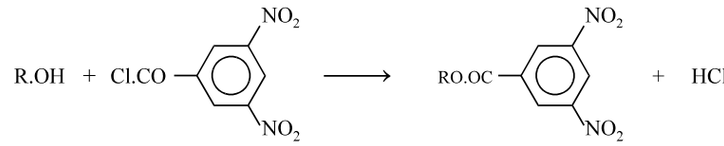
5. **Hydrogenation of the terpenoid** has also been useful in giving information regarding the carbon skeleton. Hydrogenation converts the compound into fully saturated parent hydrocarbon which may correspond to any one of the formula given below:

Formula	*Type*
C_nH_{2n+2}	Acyclic
C_nH_{2n}	Monocyclic
C_nH_{2n-2}	Bicyclic
C_nH_{2n-4}	Tricyclic
C_nH_{2n-6}	Tetracyclic etc.

6. **Functional nature of the group** should be ascertained in compounds containing oxygen.

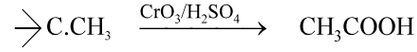
Hydroxyl group can be detected either by acetylation or by using 3,5-dinitrobenzoyl chloride



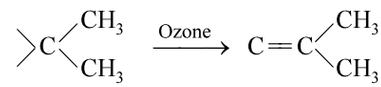
Aldehydic or *ketonic groups* can be detected by reagent like sodium bisulphite, hydroxylamine, phenylhydrazine etc.

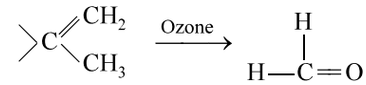
7. **Degradative oxidation** is the most powerful tool for elucidating the structure of terpenoids. Degradative oxidation gives small identifiable fragments which give valuable information regarding the structure of the compound as a whole.

Oxidation with chromic acid converts the C-methyl ($>C-CH_3$) group into acetic acid



Isopropylidene and isopropenyl groups are detected by ozone.





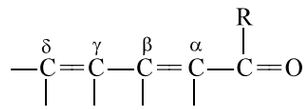
Other reagents like acid, alkaline or neutral potassium permanganate, nitric acid, lead tetra-acetate and peroxy acids are also used for oxidative degradation.

8. **Spectroscopy and other physical methods** have proved of immense value in elucidation and confirmation of the structure of terpenoids.

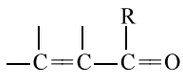
Ultraviolet spectroscopy has helped in detection and elucidation of conjugated systems. λ_{max} was observed for various terpenoids and the same was verified by a set of empirical rules developed by Woodward (1942) and modified by Fieser (1948). Woodward observed that the absorption maxima of a diene system is affected by the number and type of the substitution. The method is illustrated below:

System	λ_{max} *value*
Polyenes:	
Heteroannular (semi-cyclic) and acyclic dienes (basic value)	214 nm
Homoannular dienes (basic value)	253 nm
For each double bond extending conjugation	30 nm
For each C-substituent (alkyl gp. or ring residue)	5 nm
For each exocylic double bond	5 nm

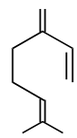
α-β unsaturated Ketones:



(R may be an alkyl group or a ring residue)

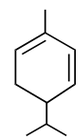
Parent system,  (basic value)	215 nm
For each double bond extending conjugation	30 nm
For each exocyclic double bond	5 nm
For each C-substituent at α – C	10 nm
For each C-substituent at β – C	12 nm
For each C-substituent at γ or δ, – C	18 nm

Some examples will illustrate the rule.



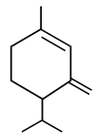
Myrcene

Observed λ_{max} 224 nm	
Calculated value:	
(*i*) For an acyclic diene	214 nm
(*ii*) For one C-substituent	5 nm
	219 nm

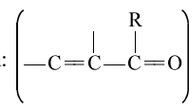


α-Phellandrene

Observed λ_{max} 263 nm	
Calculated value:	
(*i*) For homoannular diene	253 nm
(*ii*) For 3C-substituent (3 × 5)	15 nm
	268 nm



Piperitone

Observed λ_{max} 235 nm	
Calculated value:	
(*i*) For parent system: 	215 nm
(*ii*) For two substituent at β–C (2 × 12)	24 nm
	239 nm

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Infrared spectroscopy is in a way complimentary to the use of UV spectroscopy in terpenoid chemistry. Conjugated dienes and $\alpha-\beta$ ketones have nearly same λ_{max} (UV) but these can be distinguished clearly from their infrared spectra. It is useful in detecting the presence of various functional groups like hydroxyl and oxo and in distinguishing between *cis* and *trans* isomers. It is particularly helpful in detecting the presence of isopropenyl group.

Nuclear Magnetic resonance spectroscopy has also contributed in structure elucidation of terpenoids. It has helped in determining the nature of end groups and orientation of the methyl groups in the molecule. NMR spectroscopy has been helpful in determination of the number of rings and even assigning definite structures in certain case.

X-ray analysis too has been useful in elucidating the structure and stereochemistry of the terpenoids.

9. **Synthesis**. With all the physico-chemical evidence collected about the compounds a possible structure is worked out and its confirmation provided from an unambiguous synthesis.

Check Your Progress

1. Why terpenoids are sometimes also referred to as isoprenoids?
2. What are essential oils?
3. What is enfleurage process?

1.3 ISOPRENE RULE

Thermal decomposition of terpenoids give isoprene as one of the product. In 1887, Otto Wallach pointed out that terpenoids can be built up of isoprene units. This rule has been supported by following synthetic and analytical facts:

i. The empirical formula of all natural terpenoids is C_5H_8.

ii. The rule states that the terpenoid molecules are constructed of two or more isoprene units.

Fig 1.1 *Isoprene Unit*

iii. In the process of destructive distillation, terpenoids gives isoprene as main product.

iv. Isoprene undergoes polymerisation to yield various terpenoids.

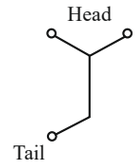
$$2C_5H_8 \xrightarrow[\text{Dimerise}]{280^\circ/\Delta} C_{10}H_{16}$$

NOTES

v. On polymerization at high temperature, isoprene gives rubber as polyterpenoid.

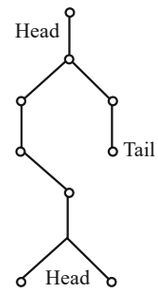
vi. In 1925 Ingold point out that a special type of arrangement were found in all terpenoids it is based on how isoprene units in terpenoids are linked together.

Special isoprene rule states that the terpenoid molecule are constructed to two or more isoprene units joined in a "head" to "tail" fashion.

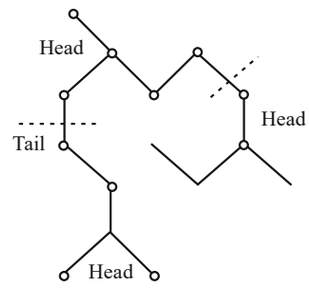


This rule is only a guiding principle and not as a fixed rule.

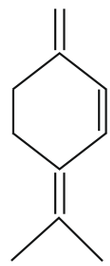
In applying isoprene rule, we look only for the skeletal unit of carbon. For example.



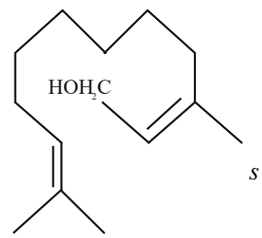
Monotrpenoid



Sesquid terpenoids



Myrcene (Monoterpene



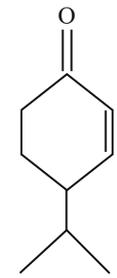
Farnesol (Sesquiter Pene)

Ingold pointed that a gem alkyl group affects the stability of terpenoids. He summarized these results in the form of a rule called "gem-dialkyl rule. Which may be stated as gem dialkyl group tends to render the cyclohexane ring unstable whereas, it stabilizes the three, four, and five member rings.

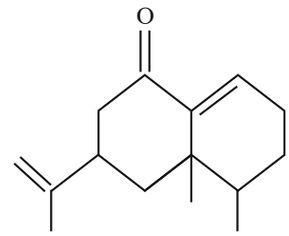
NOTES

Exception of Isoprene Rule

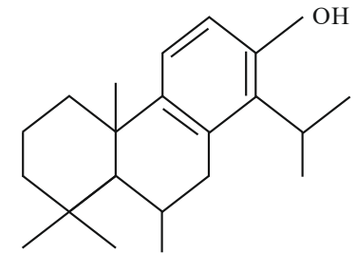
(i) Cryptone is a natural occurring terpenoid that has only nine carbon atom, hence, it does not obey the isoprene rule.



(ii) Eremophilone and Totaral do not obey the isoprene rule.



Eremophilone



Totaral

(iii) β-carotene, α-carotene, lycopene, lavandulol, etc., do not obey the isoprene rule. The isoprene units of these compounds are not liked in head to tail arrangement.

Check Your Progress

4. State the isoprene rule.
5. Write one exception of isoprene rule.
6. Which substances do not obey the isoprene rule?

1.4 SOME REPRESENTATIVE MOLECULES OF TERPENOIDS

Here we shall acquaint ourselves with some members of terpenoid family.

1.4.1 Geraniol, $C_{10}H_{18}O$

Occurrence

This terpene alcohol occurs in a large number of essential oils especially the rose, palmarosa, citronella, geranium, coriander, lemon-grass and lavender. Rose oil (obtained by steam distillation) of fresh flowers of Rosa damascena contains 40–60 per cent of geraniol. However, due to high cost, rose oil is used primarily for high grade perfumery instead of being used as a source for geraniol.

Isolation

High grade geraniol is obtained from palmarosa oil. The latter is obtained from wild-growing grass, Cymbopogon martini, by steam distillation. The oil contains 84–94 per cent of geraniol. Inferior grade geraniol is isolated from citronella oil obtained by steam distillation of Cymbopogon winterianus.

Palmarosa or citronella oil is treated with anhydrous calcium chloride. The addition compound of geraniol thus formed is then decomposed with water. An alternative method, though less satisfactory, is to crystallize its acid phthalate.

Properties

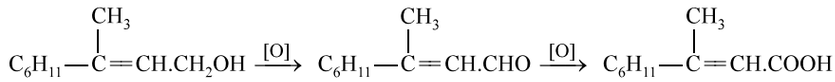
Geraniol exhibits geometrical isomerism. Geraniol is trans, while another naturally occurring alcohol nerol, present in neroli, cyclamen and bergamot oils, is *cis*-isomer.

Geraniol (*trans*) (B.P. 120°C) — Nerol (*cis*) (B.P. 225°C)

Both geraniol and nerol are colourless liquids having rose-like pleasant odour.

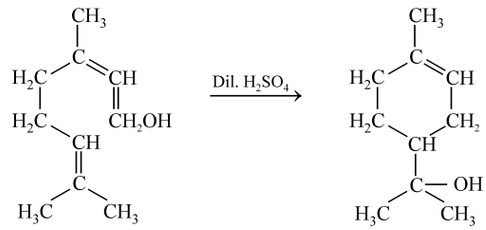
Being di-olefinic primary alcohols, both geraniol and nerol respond to reactions of carbon-carbon double bond and also of primary alcohol.

On oxidation, geraniol forms citral (geranial), which on further oxidation gives geranic acid.



Geraniol — Citral — Geranic acid

When treated with dilute sulphuric acid both geraniol and nerol form α-terpineol.



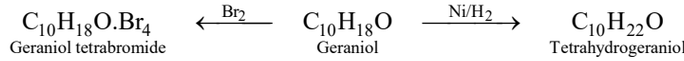
Geraniol or Nerol — α-Terpineol

Uses. Both geraniol and nerol are extensively used in perfumery, especially in the preparation of artificial rose scents.

Structure of Geraniol

1. Its molecular formula as obtained from analysis is $C_{10}H_{18}O$.

2. It contains two double bonds as shown by addition of two molecules each of hydrogen and bromine.



Geraniol tetrabromide — Geraniol — Tetrahydrogeraniol

3. On oxidation, it first forms an aldehyde (citral) and then an acid (geranic acid) both containing same number of carbon atoms. It is therefore logical to conclude that it contains a primary alcoholic group and its carbon skeleton is similar to that of citral (see later).

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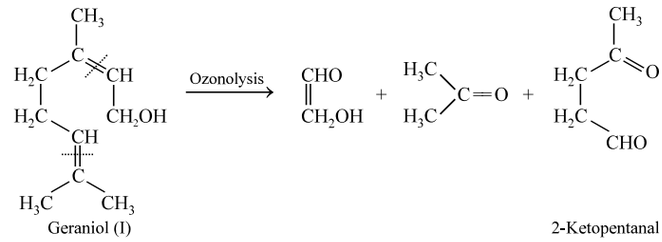
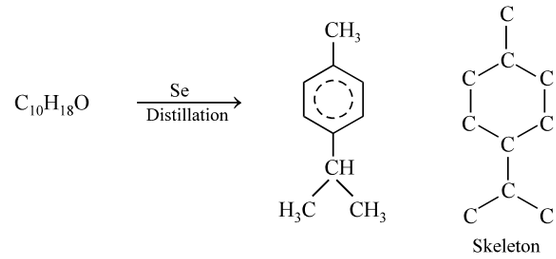
4. Ozonolysis of geraniol gave 1 mole each of glycolaldehyde, acetone and 2 ketopentanal.

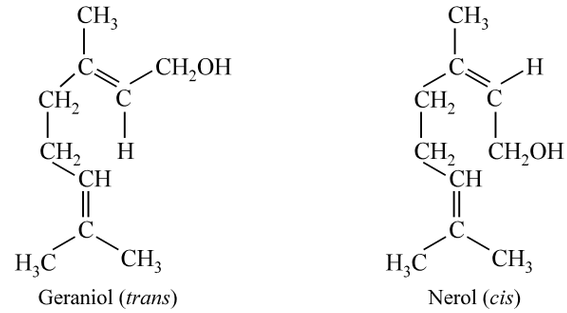


These products can be obtained if geraniol has the structure (I).

5. The nature of carbon skeleton of geraniol is confirmed by its dehydrogenation to *p*-cymene.

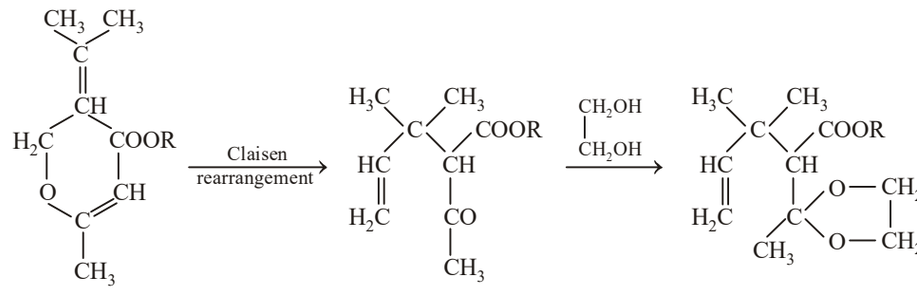


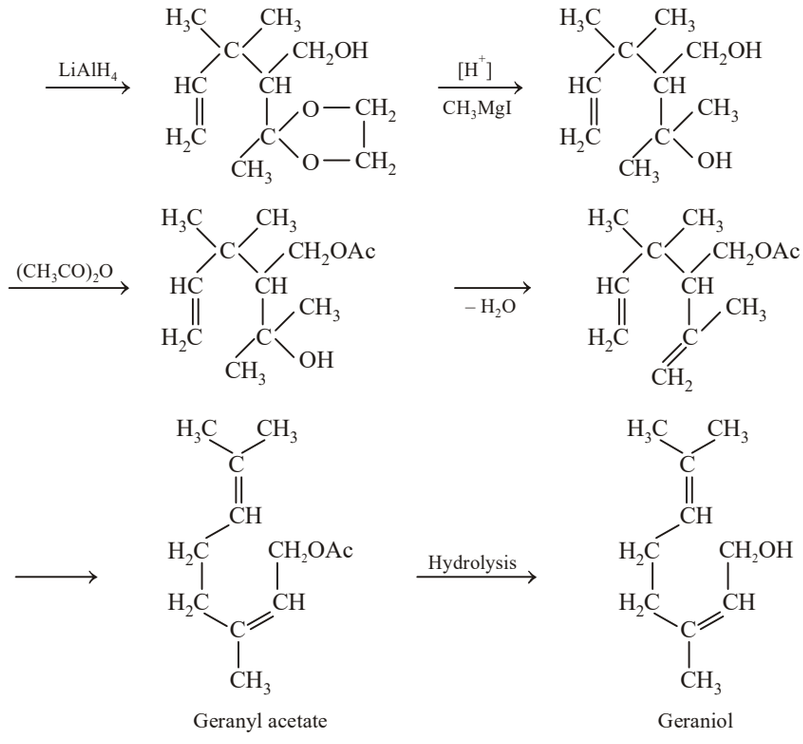
6. Keeping the above facts in view geraniol is assigned following structure.



Nerol (b.p. 225–226°C) is the geometrical isomer of geraniol. Geraniol has been assigned the *trans* configuration and nerol the *cis* because the cyclisation to α-terpeniol with sulphuric acid (see properties above) is 9 times as fast with nerol as it is with geraniol.

Synthesis. Proposed structure of geraniol has been confirmed by its synthesis.





Geranyl acetate Geraniol

1.4.2 Citral, $C_{10}H_{16}O$

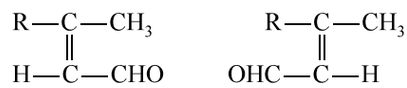
Occurrence

It is the most important open-chain terpene aldehyde. It occurs in the oil of lemon grass (70–80%) and is also present in oil of oranges, oil of lemon, oil of citronella, etc.

Isolation. It is obtained from oil of lemon grass by fractional distillation in vacuum and purified via its crystalline sodium bisulphite adduct.

Properties

Ordinary citral obtained by above method is a mixture of citral-*a* (90%) and citral-*b* (10%).



Citral-*a* (*trans*) (geranial) Citral-*b* (*cis*) (neral)

[R is $(CH_3)_2C{=}CH{-}CH_2{-}CH_2{-}$]

It is a colourless oily liquid with a strongly lemon like odour (b.p. 224–228°C d). It is optically inactive.

It is a diolefinic aldehyde and responds to reactions of olefinic linkage as well as aldehydic group. Some of its important reactions are as given below:

(i) Hydrolysis: When heated with potassium carbonate solution, cital undergoes hydrolysis forming methyl heptenone and acetaldehyde.

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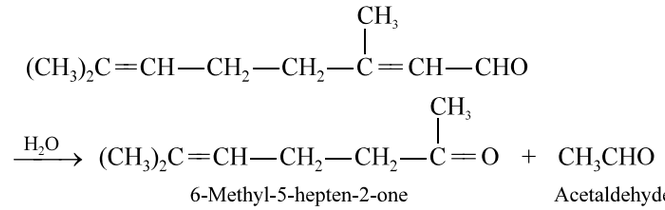
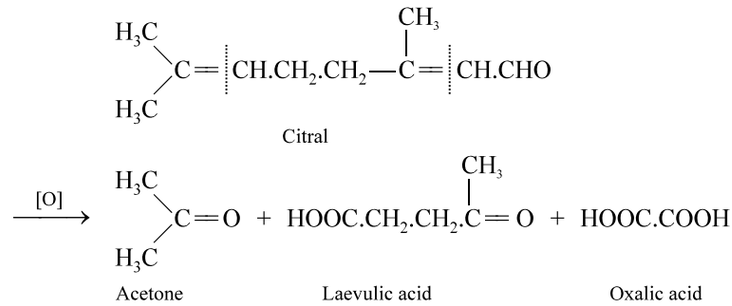


6-Methyl-5-hepten-2-one Acetaldehyde

(ii) Oxidation: Oxidation with alkaline $KMnO_4$ followed by chromic acid forms acetone, laevulic acid and oxalic acid.



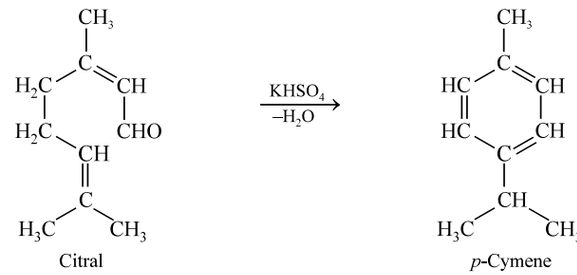
Citral Acetone Laevulic acid Oxalic acid

This gives an idea of location of double bonds in the molecule.

(iii) Dehydration: When heated with potassium hydrogen sulphate, citral loses a molecule of water forming *p*-cymene. (See structure overleaf.)

Uses. It is used:

1. As a starting material for the manufacture of α- and β-ionones.



Citral *p*-Cymene

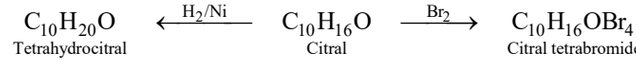
2. As a starting material for the synthesis of vitamin A.

3. In the compounding of synthetic lemon flavours.

Structure of Citral

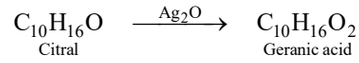
1. Molecular formula of citral as obtained from analytical data and molecular weight determination is $C_{10}H_{16}O$.

2. It has two double bonds as shown by the formation of tetrabromide and tetrahydro derivatives.



Tetrahydrocitral Citral Citral tetrabromide

3. It forms an oxime, bisulphite adduct, semicarbazone etc., and on oxidation with moist silver oxide forms geranic acid containing same number of carbon atoms. It follows, therefore, that it contains an aldehydic group.



4. On heating with potassium hydrogen sulphate citral forms *p*-cymene (Semmler, 1891)

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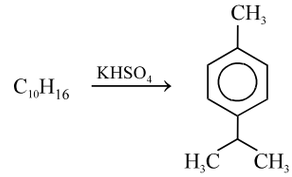
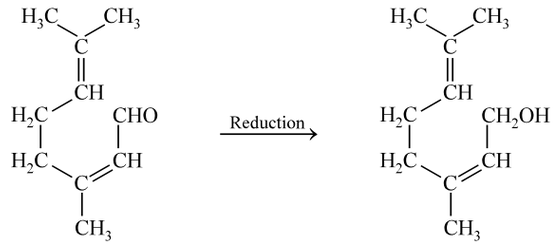
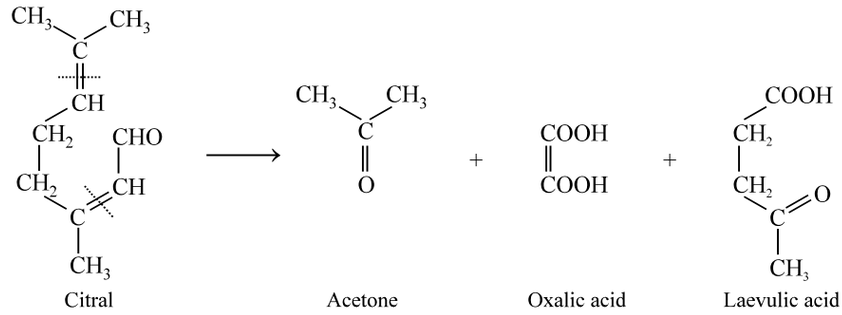


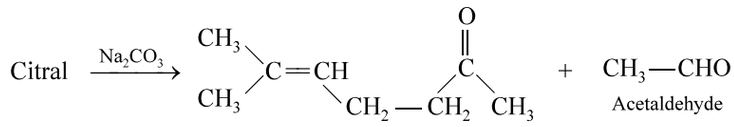
Further its complete reduction forms the hydrocarbon decane $C_{10}H_{22}$, an acyclic compound and since it could easily be converted to geraniol on reduction it was assigned an acyclic structure I by Semmler.



5. The oxidation of citral with alkaline permanganate and chromic acid gives acetone, oxalic and laevulic acids (Tiemann and Semmler, 1895). The formation of these products could be accounted for only by the assumption of above structure.



6. This structure is further supported by Verley (1897) who converted citral into methylheptenone and acetaldehyde by treatment with aqueous sodium carbonate.



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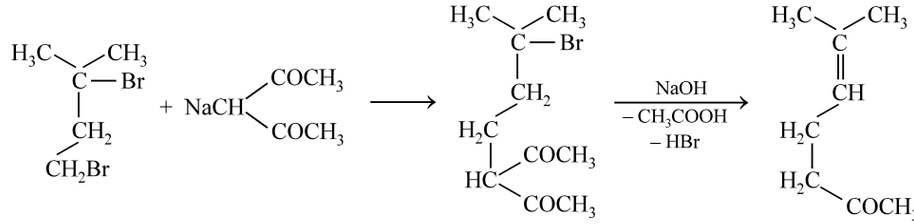
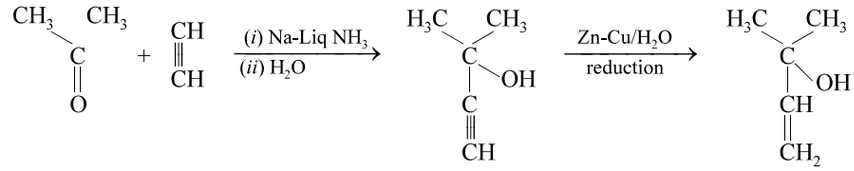
The structure of methylheptenone is known from its synthesis (Barbier and Bouveault, 1896).

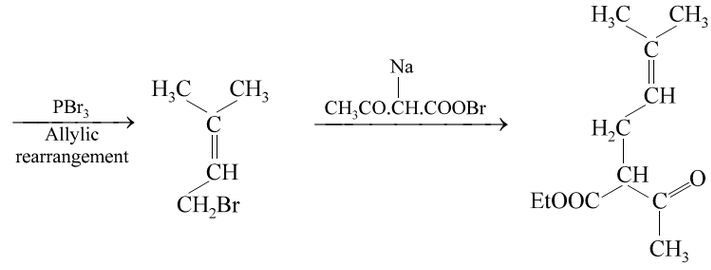


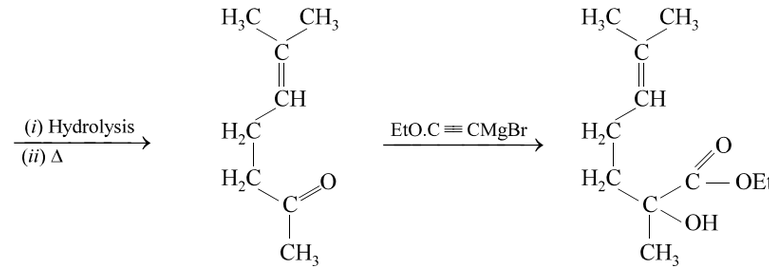
2,4-Dibromo-2-methylbutane Sodioacetylacetone Methylheptenone

7. This structure of citral is further confirmed by its synthesis.

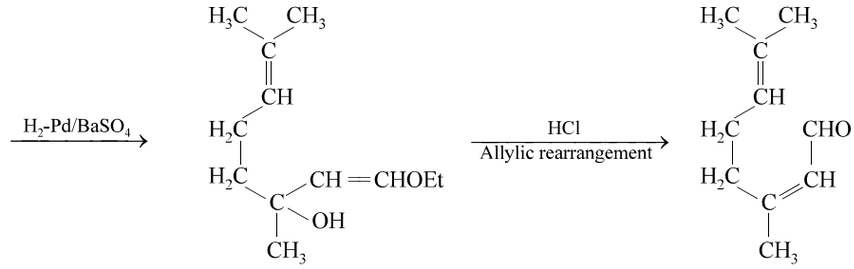
Synthesis







Methyl heptanone



Citral

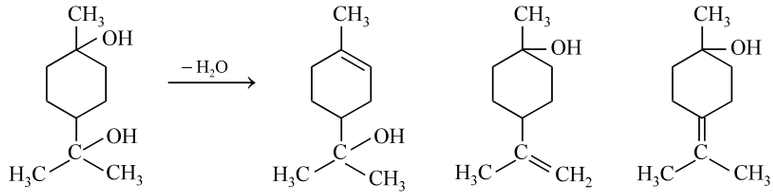
1.4.3 α-Terpineol, $C_{10}H_{18}O$

α-Terpineol is perhaps the most important monoterpenoid. It is naturally occurring optically active terpenoid whose (+), (–) and (±) forms occur in nature. The melting

pointof racemic form is 35°C and it is found in form of various esters or alone in various essential oils. The (+) form is found in petitgrain and neroli oils, the (–) form in camphor oil and the racemic form in cajeput oil.

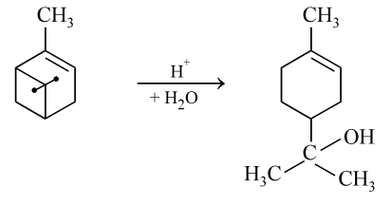
Preparation

Commercial α-terpineol for being used in cosmetics and perfumes, is obtained by dehydration of terpin which gives α, β and γ terpineols.



Terpin α-Terpineol β-Terpineol γ-Terpineol

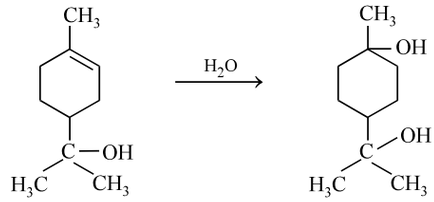
It can also be obtained by hydration of α-pinene using dilute mineral acids



β-Pinene α-Terpineol

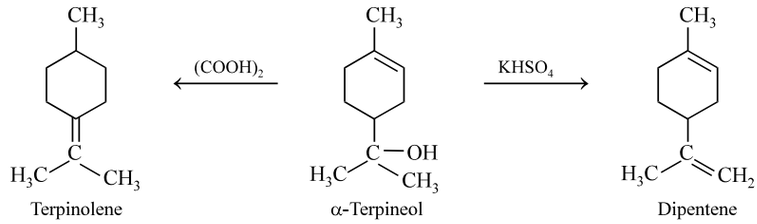
Properties

1. *Hydration* of α-terpineol gives terpin



α-Terpineol Terpin

2. Dehydration of terpineol with potassium bisulphate gives dipentene but if oxalic acid is used for dehydration terpinolene is obtained



Terpinolene α-Terpineol Dipentene

Constitution

1. Molecular formula of α-terpineol as determined by analytical data is $C_{10}H_{18}O$.

2. α-Terpineol adds two atoms of hydrogen or two atoms of bromine to give addition products showing thereby the presence of one double bond.

$$C_{10}H_{18}O.Br_2 \xleftarrow{Br_2} C_{10}H_{18}O \xrightarrow{H_2} C_{10}H_{20}O$$

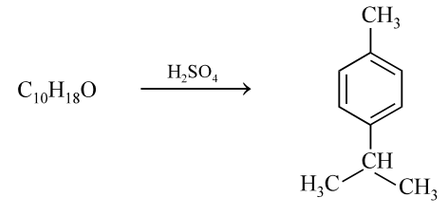
NOTES

3. α-Terpineol gets dehydrated to give an olefin which shows that alcoholic group is tertiary. Other reactions also prove that α-terpineol contains a tertiary alcoholic group.

$$C_{10}H_{18}O \xrightarrow{-H_2O} C_{10}H_{16}$$

4. From the above facts it is clear that the parent saturated hydrocarbon of α-terpineol is $C_{10}H_{20}$ ($C_{10}H_{18}O$—OH gp + 1 hydrogen atom in place of OH gp + 2 hydrogen atoms for a double bond). This formula $C_{10}H_{20}$ has the form C_nH_{2n} which represents a monocyclic system.

5. On heating with sulphuric acid, α-terpineol forms *p*-cymene in low yield. Combining this fact with the observation that α-terpineol is monocyclic, leads to the conclusion that α-terpineol has *p*-cymene carbon skeleton.



α-Terpineol *p*-Cymene

6. Thus we can conclude that α-terpineol is *p*-methane with one double bond and a tertiary alcoholic group. The positions of these functional groups was ascertained by a series of graded oxidation reaction by Wallach (1893–1895). The reactions are as given below. Only number of carbom atoms in each product are given.

$$\alpha\text{-Terpineol}\ (C_{10}H_{18}O)\ (I) \xrightarrow[KMnO_4]{1\%\ \text{Alkaline}} \text{Trihydroxy compound}\ (C_{10}H_{20}O_3)\ (II)$$

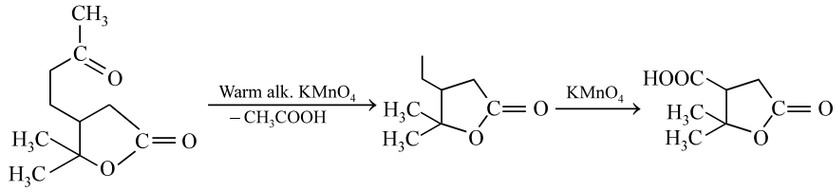
$$\xrightarrow[CH_3COOH]{CrO_3} \text{Ketohydroxy acid (Not isolated) (III)} \longrightarrow \text{Ketolactone}\ (C_{10}H_{16}O_3)\ (IV)$$

$$\xrightarrow[\text{Alk. } KMnO_4]{\text{Warmed with}} \{\text{Terpenylic acid}\ (C_8H_{12}O_4) + CH_3COOH\}_{(V)} \xrightarrow{KMnO_4} \text{Terebic acid}\ C_7H_{10}O_4\ (VI)$$

The oxidation of an α-terpineol with alkaline potassium permanganate must have hydroxylated the double bond to produce the trihydroxy compound $C_{10}H_{20}O_3$ (II). This on oxidation with chromic acid produced the compound $C_{10}H_{16}O_3$ (IV). Compound IV was neutral possessing a ketonic group. It did not react with sodium carbonate solution showing the absence of carboxyl group. However, IV on being refluxed with standard solution of sodium hydroxide, revealed that alkali has been consumed equivalent to one carboxyl group. Thus (IV) could be a lactone of the monocarboxylic acid which could not be isolated. The slow and spontaneous lactonisation indicates that the acid must have been a γ-hydroxy acid and hence IV is a γ-lactone. Oxidation of IV with alkaline potassium permanganate to yield $C_8H_{12}O_4$ (Terpenylic acid) and acetic acid points IV to be a methyl ketone having a CH_3CO— group. The nature of IV was confirmed by its synthesis (Simonsen *et. al.* 1932) and could be called as homoterpenyl-methyl ketone.

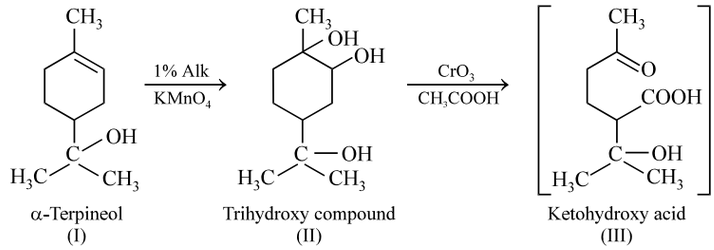
In the formation of the compound (IV) from (II) there has been no loss of carbon atoms hence the double bond must be in the ring and not outside. Had it been outside the number of carbon atoms in (IV) must have been less.

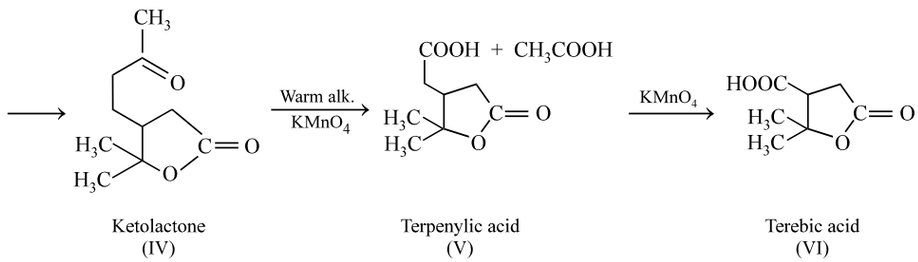
The oxidation of IV (which has been synthesised) to terpenylic acid (V) and terebic acids VI can be represented as given below.



Synthesis of terpenylic acid and terebic acid had been achieved by Simonsen (1907) which confirms their structure as V and VI respectively.

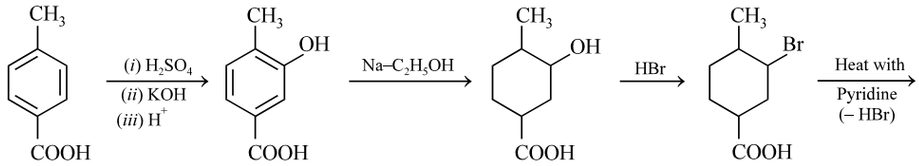
The reaction formulated above can be explained if α-terpineol is represented as *p*-menth-1-en-8-ol, the formula proposed by Wagner in 1894.

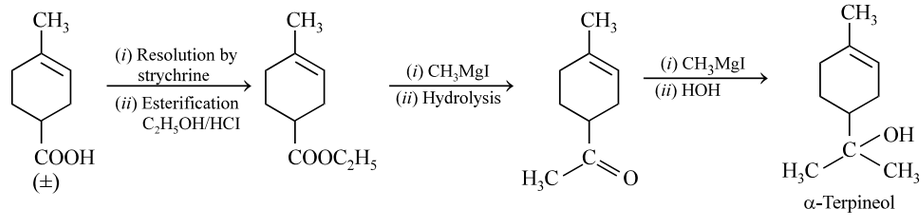




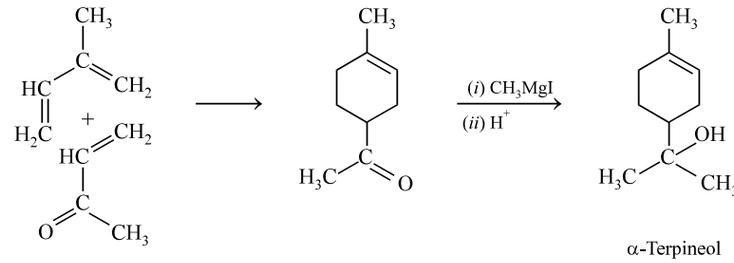
Synthesis of α-Terpineol

(*i*) First synthesis of α-terpineol was accomplished in 1904 by Perkin Junior. After four years Perkin Junior, Meldrum and Fisher synthesised α-terpineol starting from *p*-toluic acid.





(*ii*) A simpler synthesis of α-terpineol has been accomplished by Alder and Vogt (1949) using Diels-Alder reaction. Starting with isoprene and methyl vinyl ketone they synthesised α-terpineol as follows:



1.4.4 Menthol Hexahydrothymol, (p-Menthane-3-ol), $C_{10}H_{19}OH$

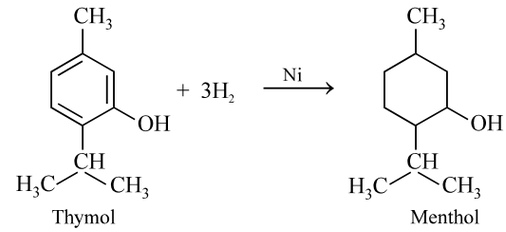
Occurrence

In its levo form menthol is found in peppermint oil (obtained from Mentha piperita) and Japanese mint oil (obtained from Mentha arvensis).

Preparation

1. **From essential oil**. Menthol is obtained by chilling essential oils. The crystals separated are pressed and purified by recrystallization.

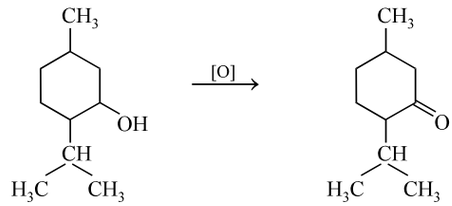
2. **From thymol**. Catalytic hydrogenation of thymol forms menthol.



Properties. Menthol forms colourless, volatile prismatic crystals (m.p. 45°C and b.p. 212°C) possessing strong peppermint odour and cooling taste. It is optically active. It exists in 8 stereoisomeric forms. Naturally occurring isomer is *l*-menthol, while the synthetic one is menthol.

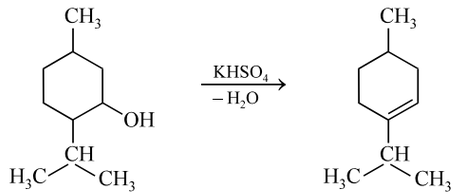
Chemically it behaves like alicyclic secondary alcohols. Some of its important reactions are discussed below:

(i) Oxidation. By chromic acid menthol is oxidised to menthone.



Menthol Menthone

(ii) Dehydration. On dehydration with potassium hydrogen sulphate menthol yields 3-menthene.



Menthol 3-Menthene

Uses

1. It is used as an antiseptic in ointments, nasal sprays, and gargles.
2. It finds use in medicine and pharmaceuticals.
3. Because of its cooling effect it is used in tooth paste and tooth powder and also is shaving creams.

1.4.5 Farnesol

Molecular formula $C_{15}H_{26}O$

Structure

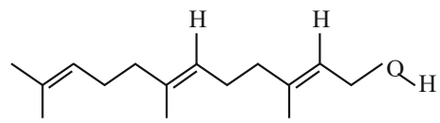


Fig. 1.2 *Farnesol*

Farnesol is farnesene sesquiterpenoid that is dodeca-2, 6, 10-triene substituted by methyl groups at positions 3, 7 and 11 and a hydroxy group at position 1. Its important sources are essential oils of ambrette seeds, citronella rose and seville orange oil. It has a role as a plant metabolite, a fungal metabolite and an antimicrobial agent.

Properties

1. Molecular weight of farnesol is 222.37.
2. Farnesol is a colourless liquid with a delicate floral odour.
3. Boiling point of farnesol is 110-113°C.

Structure Determination of Farnesol

It was determined in 1913 by Kerschbaum by using chemical method.

(i) Farnesol belongs to sesquiterpenoid class of terpenoids. It contains three isoprene units joined head to tail.

NOTES

(ii) The basic formula for calculating double bond is

DBE = (X +1) –Y/2 = 16 – 13 = 3 ($C_xH_yO_z$)

Farnesol shows presence of three double bond in the molecule

$$C_{15}H_{26}O \xrightarrow{H_2} C_{15}H_{32}O$$

Farnesol — Hexa hydrofarnesol

(iii) **Nature of Oxygen:** When farnesol is oxidized with chronic acid, farnesol (II) $C_{15}H_{24}O$ is obtained.

$$\underset{\text{Fernesol I}}{C_{15}H_{26}O} \xrightarrow{CrO_3} \underset{\text{Fernesol II}}{C_{15}H_2O}$$

(iv) **Structure of Farnesol:** Position of hydroxyl group double bond and complete structure of farnesol may be elucidated as follows

$$\underset{\text{Fernesol I}}{C_{14}H_{23}CH_2OH} \xrightarrow{CrO_3} \underset{\text{Fernesol II}}{C_{14}H_{23}CHO} \xrightarrow[(ii)\ AC_2O]{(i)\ NH_2OH}$$

$C_{14}H_{23}CN$
Cyanide Compound
III

(i) KOH
(ii) H^+

$C_{13}H_{22}O$ — V — Geronyl Ketone

$C_{14}H_{23}CO_2H$ — IV — Farnesonic acid

The following conclusions are drawn from the above reactions:

(a) Conversion of (I) farnesol to (II) farnesol by oxidation.

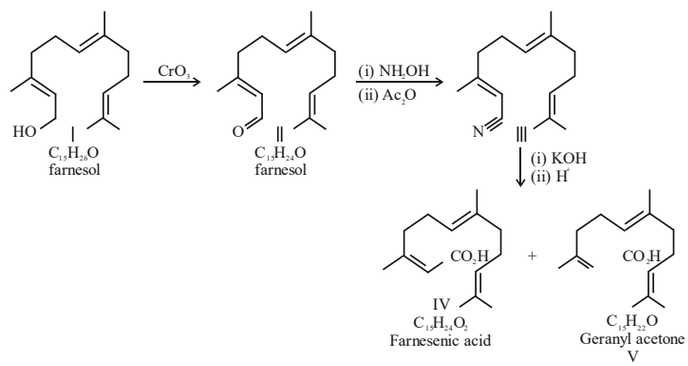
(b) Formation of (III) cyanide compound from (II) farnesol by the reaction of hydroxyl amine and AC_2O.

(c) The formation of (V) Geranyl Ketone, two carbon atoms are lost from its precursor (III) cyanide compound.

(d) The formation of farnesomic acid (Farnesoic acid) indicates the presence of α, β unsaturated carbonyl groups.

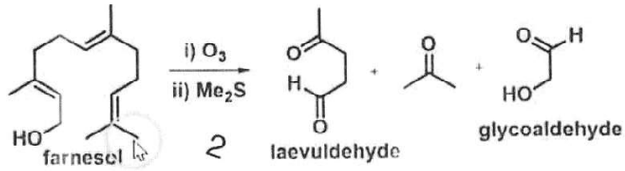
On the basis of structure of geranylacetone and reactions, the structure for farnesol (I) was given by Kerschbaum and the following facts may be formulated:

Reaction (1)



Confirmation of structure by ozonolysis of farnersol

Reaction (2)



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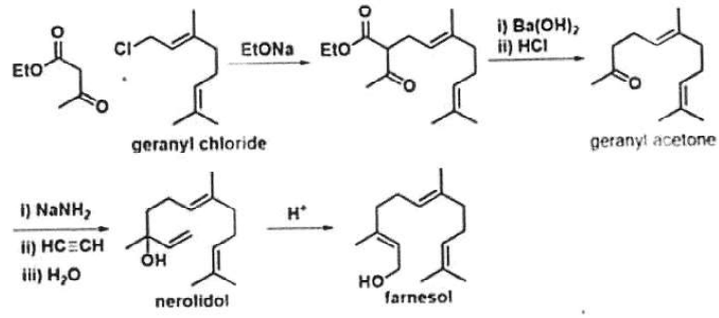
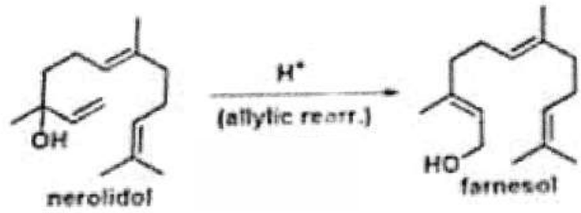
Synthesis:

(i) Synthesis of farnesol from nerolidol (Ruzicka synthesis)

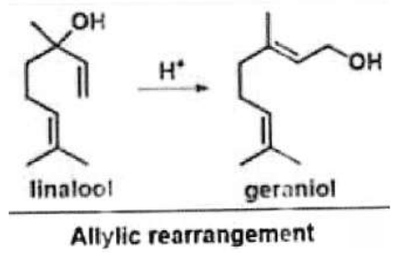
Reaction (3)



Reaction (4)

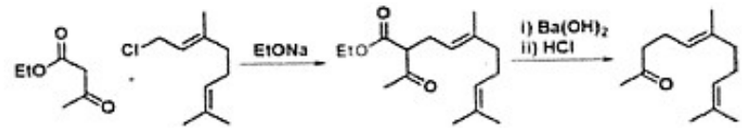


Reaction (5)



(ii) Corey synthesis:

Reaction (6)



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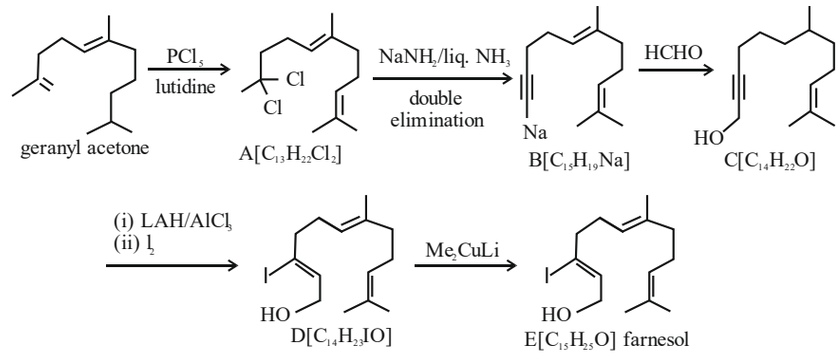
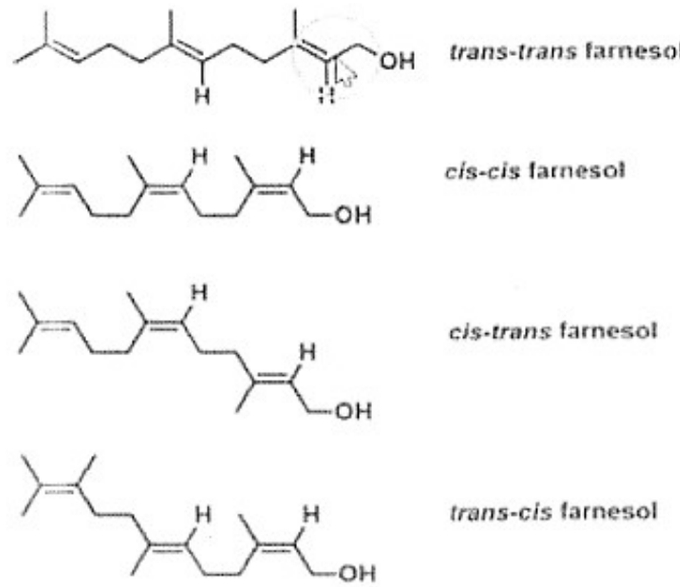


Stereoisomerism in Farnesol: In farnesol the four geometric stereoisomers are possible. In the structure *cis* and *trans* are used to indicate the position of methylene group in the main chain with respect to each other for each double bond in chain.

Reaction (7)



Uses: (i) It is used in perfumery to emphasize the fragrance of sweet floral perfumes.

(ii) Use as an essential oil.

(iii) Anti-aging skin care.

Synthesis: (F. Fischer 1928)

Farnesol on catalytic hydrogenation froms hexahydro farnesol, which in reaction with phosphorus tribromide gives hexahydrofarnesyl bromide which in react with sodium acetoacetic ester by ketonic hydralysis, from a saturated ketone. Ketone is treated with sodamide following by accetylene then is reduced with hydrogen and palladium. Reduced form phytol by the allylic rearrangement.

1.4.6 Zingiberene

Molecular formula $C_{15}H_{24}$

Structure

NOTES

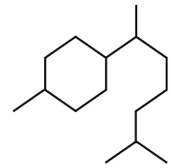


Fig. 1.3 *Structure of Zingiberene*

Zingiberene is a monocyclic sesquiterpenoids that is the predominant constituent of the oil of ginger (Zingiber officinale). From which it gets name. It can contribute upto 25-30% of the essential oils in ginger rhizomes.

Properties

(i) Boiling point of zingiberene is 134°C

(ii) It is optically active and exists in (–) from in ginger.

(1) Structure Determination of Zingiberene

On the basis of chemical method, zingiberene belongs to sesquiterpenoids class of terpenes. It contains three isoprene unit joined head to tail.

The basic formula for calculating double bond equivalence is

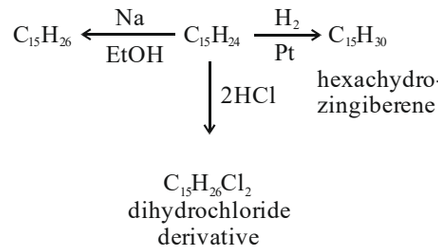
DBS = (X + 1) –Y/2 = 16 – 12 = 04

Analysis for the Nature of Double Bond in Zingiberene:

(i) On the basis of catalytic hydrogenation zingiberene takes three molecules of hydrogen to from hexahydrozingiberene.

(ii) Zingiberene is reduced by Na/EtOH to from dihydrozingiberene.

(iii) Zingiberene forms dihydrochloride derivative with HCl and thus apparently contains two double bonds.

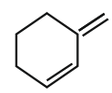


(iv) **Further Evidences for Conjugation**

(A) Zingiberene forms adduct with malic anhnydride.

(B) On the basis of UV-spectrum absorption, the λ_{max} = 260 nm (ε 2700) nm. On the above basis, the zingiberene is a homoannular diene.

Homoannulardiene
λ_{max} = 253 + ...

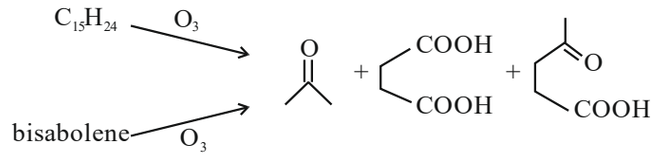


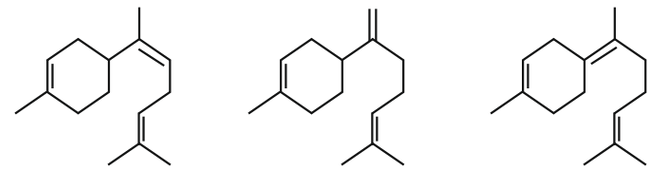
Homoannulardiene
λ_{max} = 214 + ...

NOTES

(2) Ozonolysis (Carbon Skeleton)

By ozonalysis, zingiberene gives ketone, succinic acid and laevulinic acid. Bisabolene also gives the same product on the same treatment.

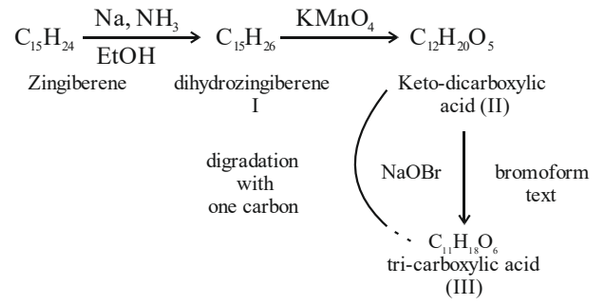


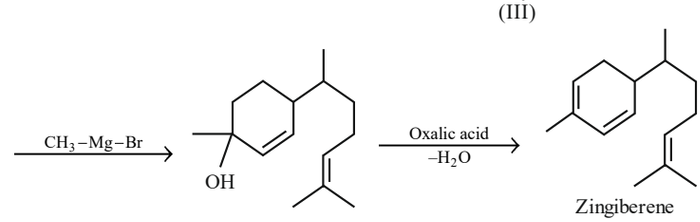


α-bisabolene β-bisabolene γ-bisabolene

(3) Oxidative Degradation of Dihydrozingiberene

Semmler (1913) suggested that zingiberene contains probably conjugated double bonds. The zingiberene can be reduced with the sodium to dihydro zingiberene and dihydro zingiberenes (I). On oxidation with potassium permanganate gives a ketodicarboxylic acid (II) ($C_{12}H_{22}O_5$) which on oxidation with sodium hypobromite from tricarboxylic acid ($C_{11}H1_8O_6$)





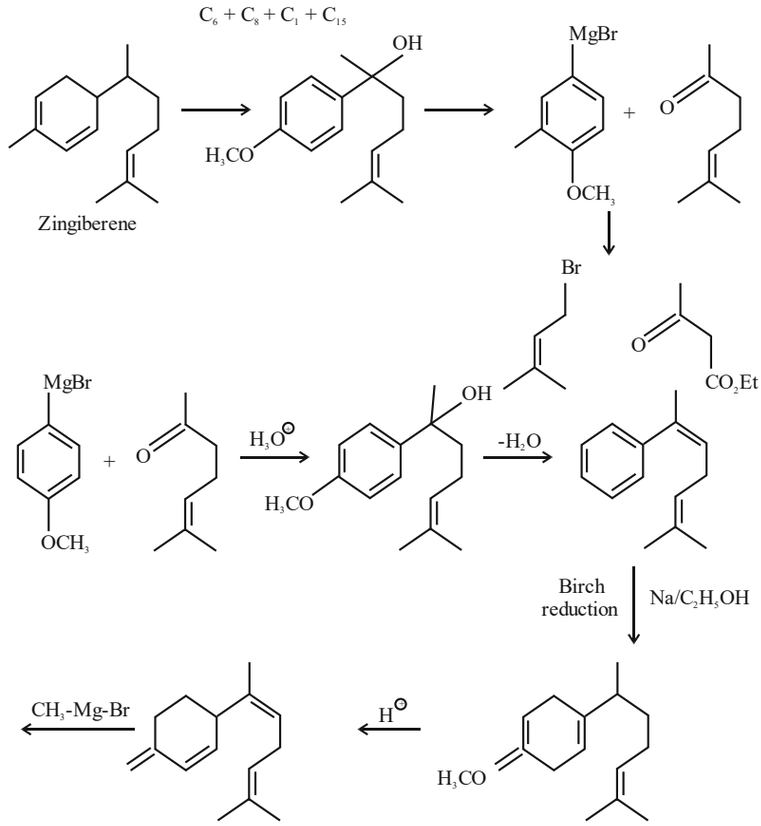
On the basis of synthesis, the zingiberene is optically active and it contains two chiral centres.

Uses: (i) It is used in cosmetics and fragrances.

(ii) It is used in antiviral, antiulcer and antifertility effects.

(4) Synthesis of Zingiberene

The structure of zingiberene is confirmed by its synthesis from ring using the birch reduction and Grignard reaction.



1.4.7 Santonin [$C_{15}H_{18}O_3$]

1. Introduction

Santonin is a sesquiterpene lactone, derived from Santonica. The unexpanded flower-heads of Artemisia maritima it is an organic solid, twining slightly yellow from the action of light and soluble in alcohol, chloroform and boiling water.

The compound was isolated in 1830 in crystalline form by extracting seeds of Artemisia cina.

2. Structure

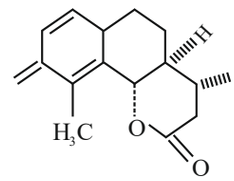


***Fig. 1.4** Structure of Santonin*

3. Structure Determination of Santonin

(i) **Molecular formula:** $C_{15}H_{18}O_3$

(ii) **Detection of Unsaturation:** Santonin contains two double bond and behave like an α-unsaturated Ketone. The presence of this group in the molecule is confirmed by its UV absorption spectrum.

$$C_{15}H_{18}O_3 \xrightarrow{2Br_2} \text{Tetrabrono Santonin}$$

NOTES

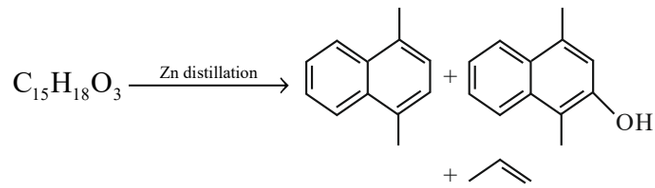
(iii) **Hydrogenation of Santonin from Tetrahydro-Santonin:** Catalytic hydrogenation of santonin forms tetrahydro-santonin ($C_{15}H_{22}O_3$), thus, confirming the presence of two double bonds.

$$C_{15}H_{18}O_3 \xrightarrow[\text{Catalytic}]{H_2} \underset{\text{Tatrahydro santonin}}{C_{15}H_{22}O_3}$$

(iv) **Nature of Oxygen (Lactone Ring):** Santonin dissolves in alkali to form the salt of the hydroxy acid, santonic acid. Hence, santonin is a lactone and its IR spectra shows a carbonyl bond at 1770 cm^{-1} and its characteristic of saturated γ-lactone.

$$\underset{\text{Santonin}}{C_{15}H_{18}O_3} \xrightarrow{NaOH} \text{hydroxy acid (Santonic acid)}$$

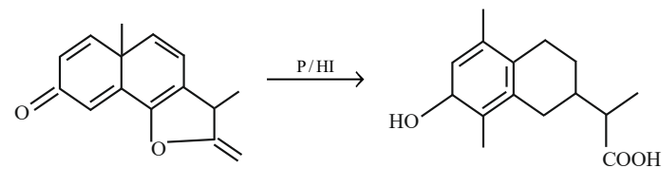
(v) **Presence of Naphthalene Skeleton:** On distillation with zinc dust, santonin gives a mixture of 1, 4 dimethylnapthaline, propene and 1, 4 dimethylnapthol.



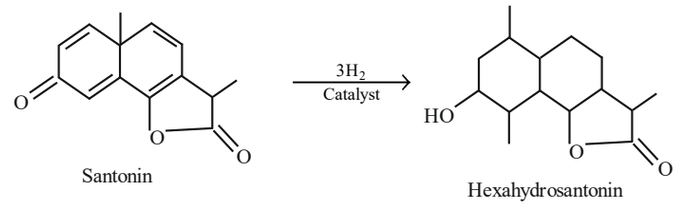
4. Confirmation of Structure of Santonin

In 1930, Clemo proposed the correct structure of santonin (I). He suggested the structure santonin, on the basis of the following reactions:

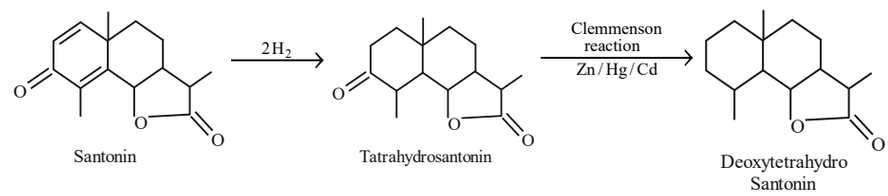
(i) Santonin(I) on reduction with phosphorous and hydraiodic acid gives santonous acid



(ii) On catalytic reduction it gives tetrahydrasantonin



(iii) On Clemmenson reduction of tetrahydrosantonin, it gives deoxytetrahydro-santonin.



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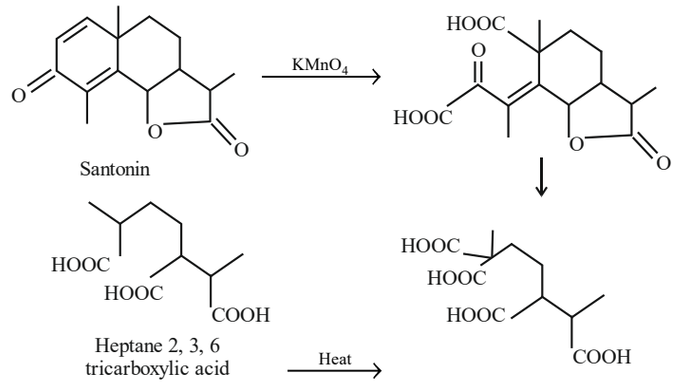
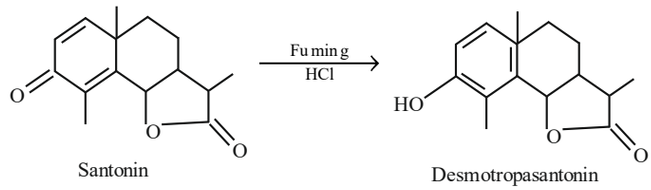
(iv) Santonin on oxidation with potassium permanganate forms tricarboxylic acid.



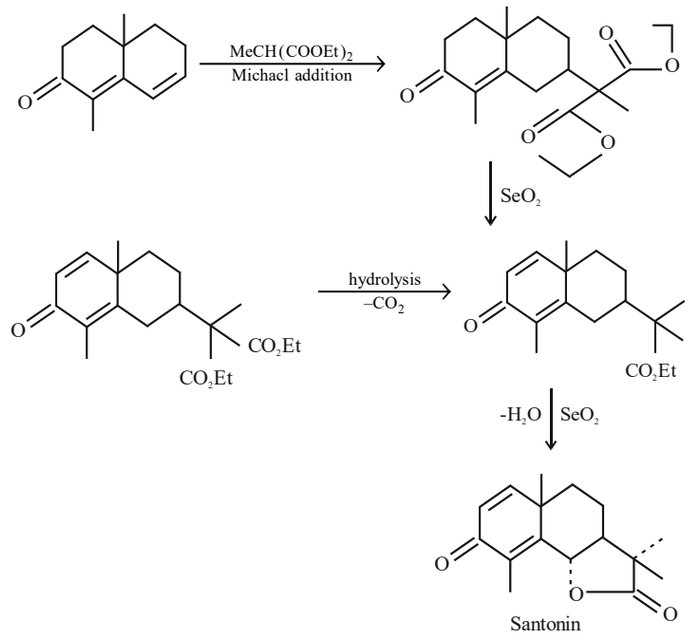
(v) Santonin on reaction with cold fuming hydrochloric acid forms desmotroposantonin.



In the above reactions, Clemo and his team agreed that if the santonin is sesquiterpenoid then it could be assumed that santonin should obey isoprene rule.

The formation of santonin acid was confirmation of isoprene rule is obeyed by the santonin.

Synthesis [ABC–1956]



Uses: (i) Santonin has anti-pyretic properties.

(ii) Santonin has anti-cancer and anti-inflammatory properties.

(iii) It is mostly used as an anti-helmintic.

NOTES

1.4.8 Phytol ($C_{20}H_{40}O$)

Phytol is an acyclic diterpenoid. It is obtained from the hydrolysis of chlorophyll. It is an optically active compound. It contains two chiral carbons.

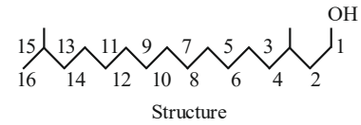


Fig. 1.5 *Structure of Phytol*

(1) Structure Determination of Phytol

(i) **Molecular Formula:** It is determined by Clemental analysis and molecular weight is $C_{20}H_{40}O$.

(ii) **Presence of Double Bond:** It is determined by the catalytic reduction. Thus, conforming the presence of one double bond.

$$C_{20}H_{40}O \xrightarrow[Pt/pd]{H_2} \underset{\text{Dihydrophytal}}{C_{20}H_{42}}$$

(iii) **Presence of Primary Alcoholic Group:** By the following reactions it can be confirmed as follow:

- **By Reaction of PCl_5:** It forms monochloro-derivatives.

$$C_{19}H_{37}CH_2OH \xrightarrow{PCl_5} \underset{\text{Phytylchloride}}{C_{19}H_{37}CH_2Cl}$$

- **By Oxidation:** It forms the same number of carbon atom compound phytanic acid.

$$C_{19}H_{37}CH_2OH \xrightarrow{[O]} C_{19}H_{37}COOH$$

- **Position of Double Bond:** On ozonolysis, phytol a saturated ketone and glycolaldehyde. (By F. Fischer 1928)

$$C_{20}H_{40}O \xrightarrow{\text{Ozonolysis}} C_{18}H_{36}O + \underset{CH_2OH}{\overset{CHO}{|}}$$

- **Acyclic Structure:** General formula C_nH_{2n+2} for acyclic compound and phytol have a fully saturated parent hydrocarbon. Hence, it must be acyclic.

- **Presence at CH_3CO Group at the End of Chain of Ketone:** By the haloform reactions, ketone contains CH_3-CO-group at the end of the chain.

$$C_{16}H_{33} - \overset{O}{\overset{\|}{C}} - CH_3 \xrightarrow{NaOBr} \underset{\text{Bromoform}}{C_{15}H_{33}COOH + CHBr_3}$$

1.4.9 Abietic Acid ($C_{20}H_{30}O_2$)

1. Introduction

(i) Abietic acid is the primary compound of resin acid is the primary irritant in pine wood.

(ii) Resin acid refers to mixture of several related carboxylic acid, primarily abietic acid, found in trees resins.

(iii) Resin acids are tacky, yellowish greens that are water insoluble.

(iv) The melting point of abietic acid is 172°C to 178°C.

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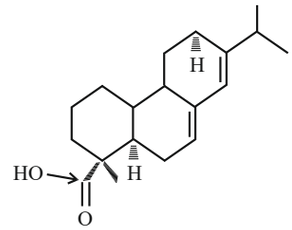


***Fig. 1.6** Structure of Abietic Acid*

2. Structure Determination of Abietic Acid

(i) **Molecular Formula:** Abietic acid is $C_{20}H_{30}O_2$.

(ii) **Presence of Double Bond:** By the catalytic hydrogenation, abietic acid converts into tetrahydro abietic acid derivative. This show the presence of the double bond.

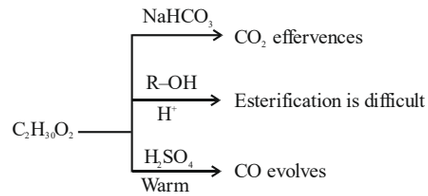
$$C_{20}H_{30}O_2 \xrightarrow[Pl]{2H_2} C_{20}H_{34}O_2$$

(iii) **Nature of Oxygen:** The presence of oxygen in the molecule may be proved by the following observation:

(A) Abietic acid gives effervescences of CO_2 with $NaHCO_3$. This shows that it contains–COOH groups.

(B) Abietic acid is difficult to esterify, therefore it may be tertiary–COOH group.

(C) Abietic acid evolves carbon monoxide with concentrated H_2SO_4, therefore it is a tertiary carboxylic acid.



(iv) **Tricyclic Nature of Abietic acid:** Parent hydrocarbon of tetrahydro-abietic acid $C_{19}H_{33}COOH$ is $C_{19}H_{34}$ which corresponds to the general formula C_nH_{2n-4} for tricyclic compounds. Hence abietic acid is tricyclic in nature.

(v) **Presence of Phenanthrene Skeleton:** Dehydrogenation of abietic acid with S or Se gives a phenanthrene derivative, $C_{18}H_{18}$ known as retene.

$$C_{19}H_{29}COOH \xrightarrow{S \text{ or } Se} \underset{\text{retene}}{C_{18}H_{18}}$$

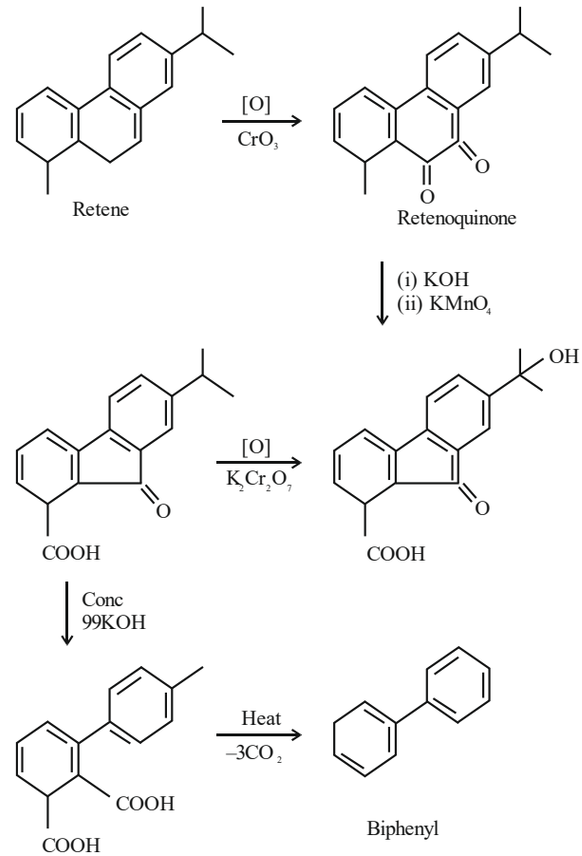
(vi) **Structure of Retene:**

NOTES

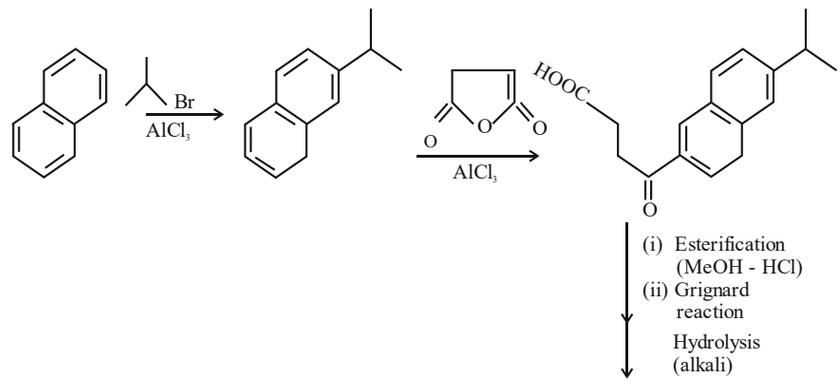
(a) Molecular formula of retene is $C_{18}H_{18}$

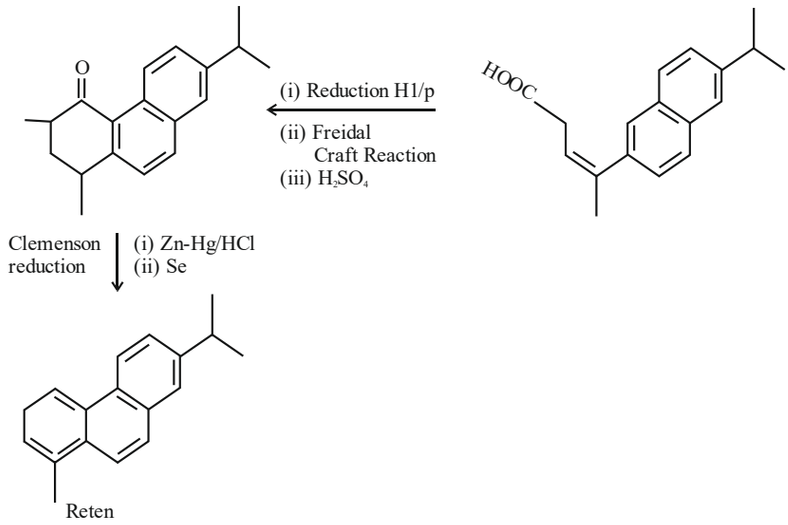
(b) By the oxidation with CrO_3, retene gives retene quinone

(c) Retene quinone on oxidation with $KMnO_4$ forms intermediate. Further oxidation with concentrated KOH, $K_2Cr_2O_7$ and heating the compound with aqueous potassium hydroxide gives biphenyl.



Haworth Synthesis of Retene

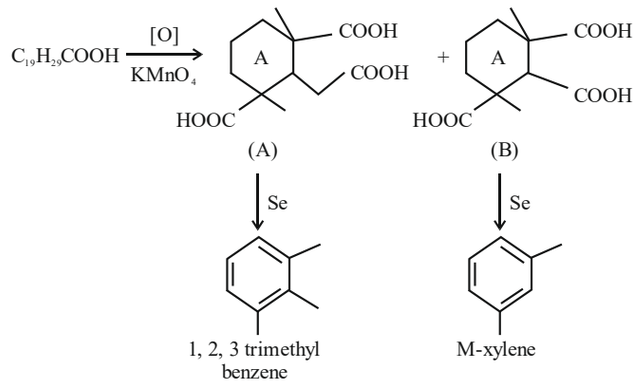




The formation of retene from abietic acid on dehydrogenation shows that the abietic acid has retene skeleton.

(5) Determination of Position of Angular Methyl Group

Abietic acid on oxidation with $KMnO_4$ gives a mixture of products, among which two are carboxylic acid, $C_{11}H_{16}O_6$ (A) and $C_{12}H_{18}O_6$ (B).



Observation

(a) Two acids (A) and (B) are derived from ring A of abietic acid.

(b) Ring B and C being unsaturated are expected to cleave during oxidation.

(c) The isolation of m-xylene and 1, 2, 3 trimethylbenzene indicates that the two methyl groups in abietic acid are located inter position with respect to each other.

Check Your Progress

7. What is the molecular formula of citral?
8. From where α- terpineol is obtained?
9. Write the physical properties of farnesol.
10. Phytol is obtained by which process?

NOTES

1.5 CAROTENOIDS

Carotenoids also called tetraterpenoids, are yellow, orange, and red organic pigments that are produced by plants and algae, as well as several bacteria, and fungi. Carotenoids give the characteristic colour to pumpkins, carrots, parsnips, corn, tomatoes, canaries, flamingos, salmon, lobster, shrimp, and daffodils. Carotenoids can be produced from fats and other basic organic metabolic building blocks by all these organisms.

There are over 1,100 known carotenoids.which can be further categorized into two classes, xanthophylls (which contain oxygen) and carotenes (which are purely hydrocarbons and contain no oxygen). All are derivatives of tetraterpenes, meaning that they are produced from eight isoprene molecules and contain 40 carbon atoms. In general, carotenoids absorb wavelengths ranging from 400 to 550 nanometers (violet to green light). This causes the compounds to be deeply colored yellow, orange, or red. Carotenoids are the dominant pigment in autumn leaf coloration of about 15-30% of tree species, but many plant colors, especially reds and purples, are due to polyphenols.

Carotenoids serve two key roles in plants and algae: they absorb light energy for use in photosynthesis, and they provide photoprotection via non-photochemical quenching. Carotenoids that contain unsubstituted beta-ionone rings (including β-carotene, α-carotene, β-cryptoxanthin, and γ-carotene) have vitamin A activity (meaning that they can be converted to retinol). In the eye, lutein, meso-zeaxanthin, and zeaxanthin are present as macular pigments whose importance in visual function, as of 2016, remains under clinical research.

The structure of carotenoids are responsible for biological abilities, including photosynthesis, photoprotection, plant coloration, and cell signaling.

The general structure of the carotenoid is a polyene chain consisting of 9-11 double bonds and possibly terminating in rings. This structure of conjugated double bonds leads to a high reducing potential, or the ability to transfer electrons throughout the molecule.

Carotenoids can transfer excitation energy in one of two ways:

(1) singlet-singlet energy transfer from carotenoid to chlorophyll, and

(2) triplet-triplet energy transfer from chlorophyll to carotenoid.

The singlet-singlet energy transfer is a lower energy state transfer and is used during photosynthesis. The length of the polyene tail enables light absorbance in the photosynthetic range; once it absorbs energy it becomes excited, then transfers the excited electrons to the chlorophyll for photosynthesis. The triplet-triplet transfer is a higher energy state and is essential in photoprotection. Light produces damaging species during photosynthesis, with the most damaging being Reactive Oxygen Species (ROS). As these high energy ROS are produced in the chlorophyll the energy is transferred to the carotenoid's polyene tail and undergoes a series of reactions in which electrons are moved between the carotenoid bonds in order find the most balanced state (lowest energy state) for the carotenoid.

Carotenoids also participate in different types of cell signalling. They are able to signal the production of absicisic acid, which regulates plant growth, seed dormancy, embryo maturation and germination, cell division and elongation, floral growth, and stress responses.

1.5.1 β-Carotene ($C_{40}H_{56}$)

1. Introduction

Beta-carotene is found in pumpkins, sweet potatoes, carrots and winter squash. β-carotene is responsible for their orange yellow colour.

Structure

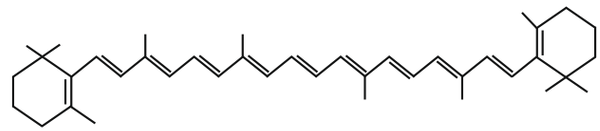


Fig. 1.7 *Structure of Beta Carotenoid*

2. Structure Elucidation of β-carotene

(i) **Molecular Formula:** Molecular formula of β-carotene is $C_{40}H_{56}$ which contain 8 isoprene units.

(ii) **Determination of Unsaturation:** By the catalytic hydrogenation of β-carotene, it is concluded that 11 double bonds are present in β-carotene.

$$\underset{\beta-\text{Carotene}}{C_{40}H_{56}} + 11H_2 \xrightarrow{Pt} \underset{\text{perhydro}-\beta-\text{carotene}}{C_{40}H_{78}}$$

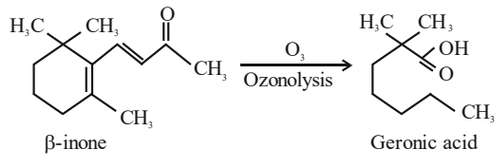
(iii) **Bicyclic Ring:** General formula C_nH2_{n-2} tends it has two rings.

(iv) **Presence of Five Conjugated Double Bonds:** The characteristic reaction of conjugated double bond, the β-carotene form adduct with 5 maleic anhydride molecules. This show that there are five conjugated double bond in β-carotene.

(v) **Ozoanalysis:** On ozonalysis, b-ionone gives one molecule of geronic acid, while β-carotene on ozonalysis gives two molecules of geronic acid. (Karror 1930).

$$\beta-\text{Carotene} \xrightarrow{air} \beta-\text{ionone}$$

$$\beta-\text{Carotene} \xrightarrow[[O]]{Cold} \beta-\text{ionone}$$



β-inone Geronic acid

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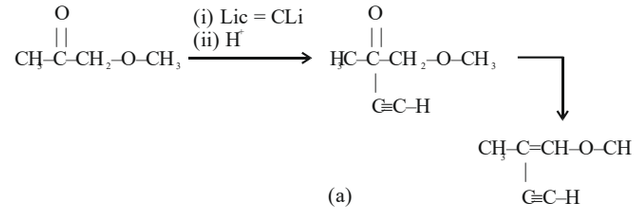
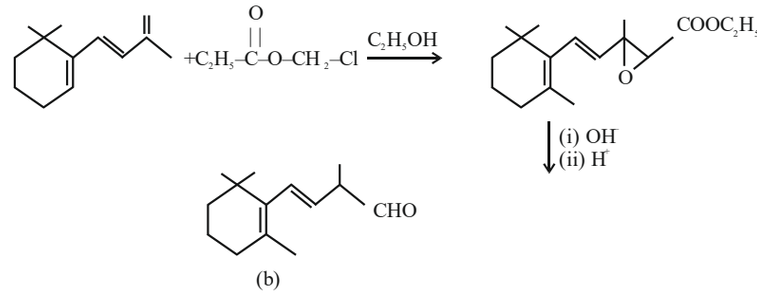
3.**Synthesis:** The structure of β-carotenes is confirmed by the following synthesis:

(A) **Inhoffen Synthesis (1950):** It involves the combination of C_{18}, C_2 and C_{19} type of skeletone. This synthesis divide in four steps:

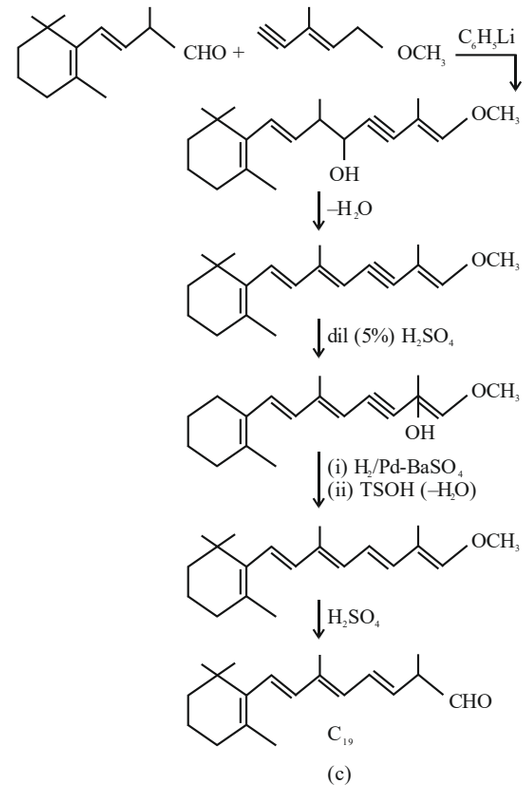
(i) Synthesis of methyl 2-methyl butenyl-3-yenl:



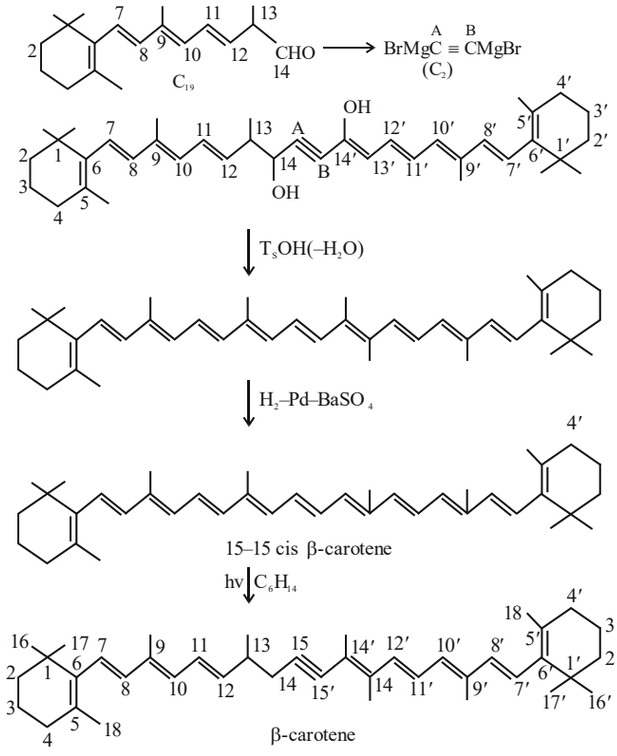
(ii) Synthesis of Moiety:



(iii) Synthesis of C_{19}:



(iv) Combination of $C_{19} + C_2 + C_{19}$: Structure



Check Your Progress

11. What are carotenoids?
12. State the role of carotenoids in plants and algae.
13. In which plants beta-carotene is found?
14. Write the molecular formula of β-carotene.

1.6 ANSWERS TO 'CHECK YOUR PROGRESS'

1. Certain fragrant substance obtained from different parts of plants, having application in perfumery, food flavouring and medicines, are known as essential oils.
2. Terpenes in general have the empirical formula C_5H_5 and are supposed to be derived from isoprene (C_5H_8). For this reason they are sometimes referred to as isoprenoids also.
3. Enfleurage process is a method applicable for the extraction of essential oils from flowers.
4. Isoprene rule states that the terpenoid molecule are constructed to two or more isoprene units joined in a "head" to "tail" fashion.

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5. Cryptone is a natural occurring terpenoid that has only nine carbon atom, hence, it does not obey the isoprene rule.
6. α-carotene, β-carotene, lycopene, lavandulol, etc., do not obey the isoprene rule
7. Molecular formula of citral as obtained from analytical data and molecular weight determination is $C_{10}H_{16}O$.
8. Commercial α-terpineol for being used in cosmetics and perfumes, is obtained by dehydration of terpin which gives α, β and γ terpineols.
9. Properties of farnesol are:
   (i) Molecular weight of farnesol is 222.37.
   (ii) Farnesol is a colourless liquid with a delicate floral fragrance.
   (iii) Boiling point of farnesol is 110-113°C.
10. Phytol is an acyclic diterpenoid. It is obtained from the hydrolysis of chlorophyll.
11. Carotenoids also called tetraterpenoids, are yellow, orange, and red organic pigments that are produced by plants and algae, as well as several bacteria, and fungi.
12. Carotenoids serve two key roles in plants and algae: they absorb light energy for use in photosynthesis, and they provide photoprotection via non-photochemical quenching.
13. Beta-carotene is found in pumpkins, sweet potatoes, carrots and winter squash.β-carotene is responsible for their orange yellow colour.
14. Molecular formula of β-carotene is $C_{40}H_{56}$ which contain 8 isoprene units.

1.7 SUMMARY

- Terpenes in general have the empirical formula C_5H_5 and are supposed to be derived from isoprene (C_5H_8).
- The terpenoids occur widely in nature in different varieties of plants.
- They are generally colourless pleasant smelling liquids, lighter than water and possessing high refractive indices.
- Degradative oxidation is the most powerful tool for elucidating the structure of terpenoids.
- Infrared spectroscopy is in a way complimentary to the use of UV spectroscopy in terpenoid chemistry.
- Thermal decomposition of terpenoids give isoprene as one of the product.
- Special isoprene rule states that the terpenoid molecule are constructed to two or more isoprene units joined in a "head" to "tail" fashion.
- High grade geraniol is obtained from palmarosa oil.
- On oxidation geraniol forms citral (geranial), which on further oxidation gives geranic acid.

NOTES

- Citral is the most important open-chain terpene aldehyde. It occurs in the oil of lemon grass.
- Molecular formula of citral as obtained from analytical data and molecular weight determination is $C_{10}H_{16}O$.
- α-Terpineol is perhaps the most important monoterpenoid. It is naturally occurring optically active terpenoid whose (+), (–) and (±) forms occur in nature.
- In its levo form menthol is found in peppermint oil (obtained from Mentha piperita) and Japanese mint oil (obtained from Mentha arvensis).
- It is also present in oil of oranges, oil of lemon and oil of citronella etc.
- Farnesol is farnesane sesquiterpenoid that is dodeca-2, 6, 10-triene substituted by methyl groups at positions 3, 7 and 11 and a hydroxy group at position 1.
- In farnesol, the four geometric stereoisomers are possible.
- Zingiberene is a monocyclic sesquiterpenoids that is the predominant constituent of the oil of ginger (Zingiber officinale).
- On the basis of chemical method, zingiberene belongs to sesquiterpenoids class of terpenes.
- Santonin is a sesquiterpene lactone, derived from Santonica.
- Phytol is an acyclic diterpenoid. It is obtained from the hydrolysis of chlorophyll.
- Abietic acid is the primary compound of resin acid is the primary irritant in pine wood.
- The formation of retene from abietic acid on dehydrogenation shows that the abietic acid has retene skeleton
- Carotenoids also called tetraterpenoids, are yellow, orange, and red organic pigments that are produced by plants and algae, as well as several bacteria, and fungi.
- The general structure of the carotenoid is a polyene chain consisting of 9-11 double bonds and possibly terminating in rings.
- Beta-carotene is found in pumpkins, sweet potatoes, carrots and winter squash. β-carotene is responsible for their orange yellow colour.
- Molecular formula of β-carotene is $C_{40}H_{56}$ which contain 8 isoprene units.

1.8 KEY TERMS

- **Essential Oils:** Certain fragrant substance obtained from different parts of plants, having application in perfumery, food flavouring and medicines, are known as essential oils.
- **Enfleurage Process:** Enfleurage process is a method applicable for the extraction of essential oils from flowers.

NOTES

- **Farnesol**: Farnesol is farnesane sesquiterpenoid that is dodeca-2, 6, 10-triene substituted by methyl groups at positions 3, 7 and 11 and a hydroxy group at position 1.
- **Santonin:** Santonin is a sesquiterpene lactone, derived from Santonica.
- **Abietic Acid**: Abietic acid is the primary compound of resin acid is the primary irritant in pine wood.
- **Carotenoids**: Carotenoids also called tetraterpenoids, are yellow, orange, and red organic pigments that are produced by plants and algae, as well as several bacteria, and fungi.
- **Beta-Carotene**: Beta-carotene is found in pumpkins, sweet potatoes, carrots and winter squash. b-carotene is responsible for their orange yellow colour.

1.9 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. Write the method of isolation of terpenoids.
2. Write the classification of terpenoids.
3. Give a few examples of some important classes of terpenoids
4. What is the exception of isoprene rule?
5. Write the properties of geraniol.
6. What are the uses of citral?
7. State the moleculer formula of alpha terpineol.
8. What is the stereoisomerism in farnesol?
9. State about ozonalysis of zingiberene.
10. What are caratonoids?
11. How the structure elucidation of β-carotene is done?

Long-Answer Questions

1. Explain the general properties of terpenoids.
2. Describe the general procedure for determining the structure of terpenoids
3. Elaborate on the synthetic and analytical facts that support isoprene rule.
4. Explain the structure of geraniol.
5. Analyse the properties of menthol.
6. Explain the structure determination of farnesol.
7. Explain the role of carotenoids in plant.

1.10 FURTHER READING

Krishnaswamy, N. R. 1999. *Chemistry of Natural Products A Unified Approach*. Himayatnagar, Hyderabad: Universities Press.

Bhat, Sujata V., B.A. Nagasampagi, Meenakshi Sivakumar · 2005. *Chemistry of Natural Products*. Narosa Publishing House

Rahman,Atta-ur. 2020. *Studies in Natural Products Chemistry.* Volume 67. Amsterdam, Netherlands: Elsevier Science.

Cooper, Raymond. , George Nicola. 2014. *Natural Products Chemistry-Sources, Separations and Structures.* London UK: Taylor & Francis.

NOTES

UNIT 2 ALKALOIDS

NOTES

Structure

2.0 INTRODUCTION

Alkaloids are a class of basic, naturally occurring organic compounds that contain at least one nitrogen atom. This group also includes some related compounds with neutral and even weakly acidic properties. Some synthetic compounds of similar structure may also be termed alkaloids. In addition to carbon, hydrogen and nitrogen, alkaloids may also contain oxygen, sulphur and, more rarely, other elements such as, chlorine, bromine, and phosphorus Alkaloids constitute a structurally diverse array of natural products, and these compounds have a wide range of biological activities. Many have important pharmaceutical uses. Plants are regarded as the oldest source of alkaloids, and some of the most widely recognized alkaloids, such as, morphine, quinine, strychnine, and cocaine, are derived from plants. Most alkaloids contain oxygen in their molecular structure; those compounds are usually colourless crystals at ambient conditions. Oxygen-free alkaloids, such as nicotine or coniine, are typically volatile, colourless, oily liquids. Most alkaloids are weak bases, but some, such as, theobromine and theophylline, are amphoteric. Most alkaloids have a bitter taste or are poisonous when ingested. Alkaloid production in plants appeared to have evolved in response to feeding by herbivorous animals; however, some animals have evolved the ability to detoxify alkaloids. Some alkaloids can produce developmental defects in the offspring of animals that consume but cannot detoxify the alkaloids.

In this unit, you will study about definition, nomenclature, physiological action, occurrence, isolation of alkaloids, general methods of structure elucidation of alkaloids, degradation in alkaloids, classification of alkaloids, and the role of alkaloids in plants.

2.1 OBJECTIVES

NOTES

After going through this unit you will be able to:

- Give definition, nomenclature physiological action, occurrence, isolation of alkaloids
- Analyse general methods of structure elucidation of alkaloids
- Explain degradation in alkaloids
- Describe classification of alkaloids
- Comprehend the role of alkaloids in plants

2.2 BASIC CONCEPT OF ALKALOIDS

The term *alkaloid*, meaning *alkali like*, usually applies to basic nitrogenous susbtances of plant origin, with marked physiological action. The basic character may be due to amino group, imino group or nitrogen atom in a heterocyclic system. The definition is, however, not perfect. *Piperine*, an alkaloid of pepper, is neither basic nor has any physiological action. Not only that a number of naturally occurring open-chain bases with marked physiological activity, *e.g.*, cholines, amino acid, phenylethyl amine etc., are referred to as vegetable bases rather than alkaloids. There are many synthetic compounds, which are closely related to alkaloids in structure and physiological action but are not considered as alkaloids *e.g.*, *adrenaline* closely resembles the alkaloid *ephedrine*, but is not referred to as an alkaloid. Similarly caffeine, which is basic and a heart stimulant and diuretic, is better referred to as purine derivative rather than an alkaloid.

Many of these alkaloids, *e.g.*, *cocaine* (local anaesthetic), *morphine* (analgesic), *quinine* (antimalarial), *atropine* (for dilating pupil of the tye), *reserpine* (for lowering blood pressure) etc., are useful drugs. Though the therapeutic and poisonous properties of various plants were known in early times, the first alkaloid, morphine, was isolated by Serturner only in 1817 from opium. Later on *strychnine, brucine, quinine* and many other alkaloids were isolated. All attempts to synthesise and to determine the constitution of alkaloids remained a serious problem for scientists for a long time. It was only in 1870 and onwards that the pioneer work of synthesis and determination of their structure was done by Skraup, Willstatter, Perkin (junior), Robinson and others.

2.2.1 Nomenclature

Structurally alkaloids have great diversity, so no convenient and systematic method is possible for naming them. However, they are named according to (*a*) *the name of chief investigator e.g.*, pelletierine (after Pelletier), (*b*) *botanical sources e.g.*, papaverine (from *Papaver somniferum*), coniine (from *Conium maculatum*), (*c*) *physiological action, e.g.*, emetine, morphine, etc. Prefixes such as, *epi., iso., neo., pseudo*, etc., or Greek letters are often used to designate isomeric alkaloid or the one with slightly modified structure. The prefix *nor*- is used to denote a structure, which has no methyl group attached to nitrogen atom.

2.2.2 Occurrence

Alkaloids occur chiefly in dicotyledonous plants as salts of common plant acids such as, malic, citric, oxalic, succinic or tannic and sometimes of special acids like quinic acid in the alkaloid of cinchona bark and meconic acid in those of morphine group. Rarely they occur as glycosides also, *e.g.*, solanine. More than two thousand alkaloids are known which occur in forty different plant families. However, they are rarely found in lower plants like algae and fungi etc.

NOTES

Although alkaloids may be detected in all parts of plant, yet they accumulate to a greater extent in fruits, seeds and sometimes also in bark. Alkaloid content of the plant varies with the species and age of the plant. Seasonal, cultural and climatic variations do affect the content, though to a lesser extent. Many different alkaloids, usually related in structure, may be present in the same species, *e.g.*, opium poppy contains about 20 closely related alkaloids. A particular alkaloid is rarely present in different plant families.

2.2.3 Extraction/Isolation

Isolation and purification of alkaloids from plant materials is a considerably laborious task, because of the presence of several closely related compounds. In addition to that the presence of plant products like glycosides, organic acids, etc., make the task still more difficult. However, a general outline is as given below.

The dried plant material is extracted with petroleum ether and filtered to remove fat. The residue is extracted with boiling methanol and filtrate so obtained is evaporated. The residual mass is lixiviated with acidulated water, containing hydrochloric or sulphuric acid, to extract base as soluble salt. The suspended impurities are removed and the filtrate extracted with chloroform or ether to remove water soluble material. The extract is steam distilled to remove steam volatile alkaloids and rest of the solution made alkaline with sodium carbonate, ammonium hydroxide or lime to liberate free bases. The bases are again extracted by using ether or chloroform. The mixture of the bases thus obtained is then separated into components by methods like fractional crystallisation, fractional precipitation and various chromatographic techniques.

A recent method developed by Lee (1960) consists in treating plant alkaloid with Reinecke's solution [$H\{Cr(NH_3)_2(SCN)_4\}$] to convert the alkaloid into reineckates. These are dissolved in acetone and the alkaloids obtained in a state of purity by passing the solution through an ion exchange column.

2.2.4 General Characteristics

(*i*) **State**. Alkaloids are generally colourless crystalline solids, which cannot be distilled. A few of them *e.g.*, nicotine and coniine are liquids and volatilize without decomposition.

(*ii*) **Solubility**. Except liquid alkaloids others are either insoluble or sparingly soluble in water, but dissolve more or less in chloroform, ether or benzene. However, they are readily soluble in alcohol.

(*iii*) **Optical Activity**. They are optically active, the majority being laevorotatory.

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(*iv*) **Basic Nature**. Being strongly basic they form salts both with inorganic and organic acids *e.g.* hydrochloric, sulphuric, perchloric, oxalic, tartaric, citric, salicylic acids etc. Unlike parent alkaloids their salts are generally soluble in water, but insoluble in organic solvents. Their chlorides, sulphates and oxalates readily crystallize. Their chlorides form double salts with auric and platinic chlorides.

(*v*) **Precipitation**. From their aqueous or acid solution they can be precipitated by a number of substances known as alkaloid reagents, the common ones being picric acid (*Mayer's reagent*), tannic acid, perchloric acid, potassium mercuric iodide (*Mayer's reagent II*), potassium bismuth iodide (*Dragendroff's reagent*), phosphomolybdic acid and phosphotungstic acid. These reagents form precipitates with characteristic colours and definite composition and are very sensitive. So these may be used for the detection of alkaloids even when present in minute concentration. However, they cannot be used for quantitative estimation of alkaloids because the precipitate is not sufficiently insoluble and the reagents precipitate other organic substances also. Nitric acid (*Erdmann's reagent*), Molybdic acid (*Forhde's reagent*) and formalin (*Marquis reagent*) give specific colours with alkaloidal salts.

(*vi*) **Physiological Action**. Most of the alkaloids have intensely bitter taste and often have marked physiological action when administered to animals.

Check Your Progress

1. Define the term alkaloid.
2. Where do the alkaloids generally occur?
3. What is the physical state of alkaloids?
4. What is the basic nature of alkaloids?

2.3 GENERAL METHOD FOR STRUCTURE OF ELUCIDATION OF ALKALOIDS

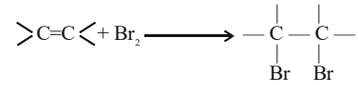
The following steps are used to establish the molecular structure of alkaloids.

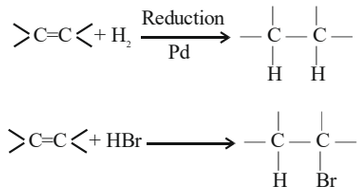
1. Molecular Formula

With qualitative and quantitative analysis, empirical formula of the compound is determined. Molecular weight is determined by physical methods or by mass spectrometry. From that molecular formula of the compound is determined. If alkaloid is optically active, its specific rotation is also measured.

2. Unsaturation Determination

Presence of unsaturation or double bond is determined by addition reaction with Br_2, H_2 or with halos acids.





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3. Nature of Oxygen

If the alkaloid contains oxygen atom then it may be in the form of –OH, C=O, –COOH, $–OCH_3$, $–CONH_2$, R–O–R', –COOR etc.

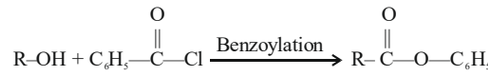
(A) **Hydroxyl Group (–OH):** The presence of hydroxyl group is established by the following reactions

(i) **Acetylation:** In this reaction acetic anhydride, acetyl chloride react and form acetyl derivative.

$$R-OH + \underset{\text{Acetyic anhydride}}{(CH_3CO)_2O} \xrightarrow{\text{Acetylation}} R-O-CO-CH_3 + CH_3COOH$$

$$R-OH + \underset{\text{Acetyic Chloride}}{CH_3COCl} \xrightarrow{\text{Acetylation}} R-O-CO-CH_3 + HCl$$

(ii) **Benzoylation:** Hydroxyl group react with Benzoyl chloride it forms benzoyl derivative.



(B) **Phenolic Group (–OH):** Phenolic group in the molecule is determined by

(i) **Solubility:** alkaloid is soluble is alkali

(ii) The compound is soluble in NaOH solution and it gives violet or green colour with $FeCl_3$ solution.

(C) **Alcoholic Group (–OH):** The presence of alcoholic hydroxyl group is confirmed by the following reactions:

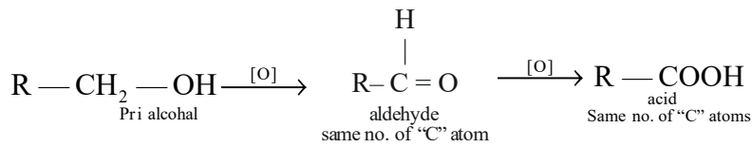
(i) It shows dehydration reaction with conc. H_2SO_4 or with P_2O_5

(ii) It forms halide with thionyl chloride

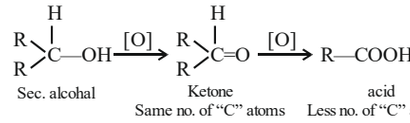
$$R–OH + SOCl_2 \rightarrow R–Cl + SO_2 \uparrow + HCl \uparrow$$

(iii) Alcoholic hydroxyl group may be primary, secondary or tertiary they are distinguished by oxidation reactions of alcohols.

Primary Alcohol: Primary alcoholic group on oxidation gives an aldehyde and then acid with the same number of carbon atom as the original molecule.



Secondary Alcohol: By the oxidation of secondary alcohol gives a ketone. The ketone on further oxidation gives a mixture of acid with lesser number of carbon atoms.



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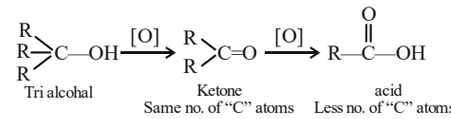
Tertiary Alcohol: Tertiary alcohol on oxidation gives a less number of ketone and acids.



(D) **Carboxylic Acid Group (–COOH):** It's determined by the solubility of alkaloid in aqueous sodium carbonate or ammonia. The number of –COOH groups in alkaloid is determined by the acid, alkali titration or by silver salt method.

(E) **Carbonyl Group:** It's determined by the formation of oxime, semicarbazone and phenylhydrazone with hydroxyl amine, semicarbazide and phenylhydrazine successively.

(F) **Ester (–COOR), Amide ($-CONH_2$) and Lactone [R-CH-CH_2-CH_2-CO-O]:** These groups are determined by hydrolysis with water, acid or alkali.

$$-COOR \xrightarrow[\text{Hydrolysis}]{NaOH} COONa + R - OH$$

$$-CONH_2 \xrightarrow[\Delta]{NaOH} R - COONa + NH_3$$

$$R - CH - CH_2.CH_2 - CO - O \xrightarrow[\Delta]{NaOH} R - \underset{\displaystyle OH}{\underset{|}{CH}} - CH_2 - CH_2 - COONa$$

(G) **Methoxy Group:** Alkaloid is heated with concentrated solution of HI at 126°C. The methoxy groups are convert into methyl iodide. Methyl iodide is absorbed by silver nitrate, when silver iodide is precipitated.

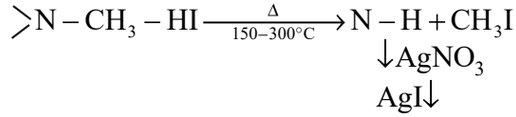
$$—OCH_3 \xrightarrow[126°C]{HI} CH_3I + OH^+$$
$$\downarrow AgNO_3$$
$$AgI\downarrow$$

(H) **Methylene Di-oxy Group (–O–CH_2–O–):** it's determined by the formation of formaldehyde, when the alkaloid is treated with HCl or H_2SO_4.

4. Nature of Nitrogen

The presence of N-methyl group is often detected by distillation of amine with soda lime or estimated by the treatment with HCl at 150–300°C and conversion of methyl iodide produced to silver iodide as mentioned for estimation of methoxy group.

$$\rangle N - CH_2 + CaO \xrightarrow[150-300°C]{NaOH/\Delta} CH_3 - NH_2$$



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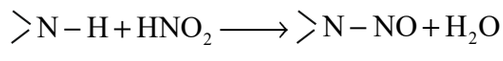
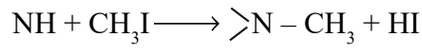
The acetylation or benzoylation can distinguish tertiary amine from secondary amine, the former being most whereas the latter gives acetate or benzoate derivative.

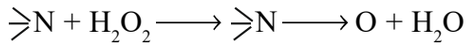
It can also done by the reaction with HNO_2 or oxidation with 30% H_2O_2

Secondary amine





Tertiary amine



Check Your Progress

5. How presence of unsaturation or double bond is determined in alkaloids?
6. What happen when alkaloid is heated with concentrated solution of HI at 126°C?
7. How the presence of N-methyl group is often detected?

2.4 DEGRADATION OF ALKALOIDS

1. **Hofmann Exhaustive Methylation:** In this method, the alkaloid is first hydrogenated and converted into quaternary methyl ammonium hydroxide, which on heating loses a molecule of water. The hydroxyl group is eliminated from tetra methyl ammonium hydroxide and the hydrogen atom from the beta-position with respect to the N atom resulting in ring opening at the "N" atom on the same side from which the β hydrogen was eliminated. The process is repeated on the formed product till the N is eliminated and an unsaturated hydro carbon is left which isomerizes to a conjugated diene.

   The Hoffmann's exhaustive methylation fails in cases where the alkaloids do not contain a β-hydrogen atom.

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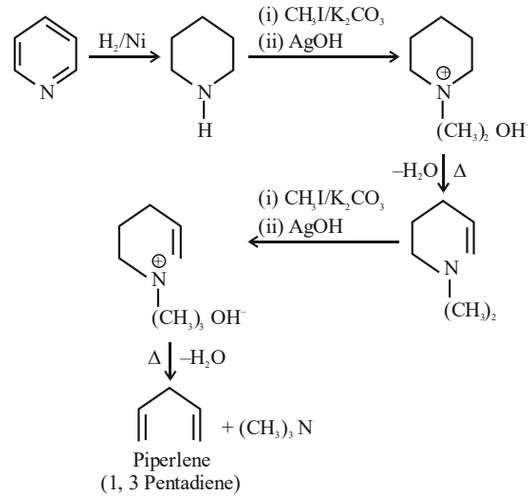
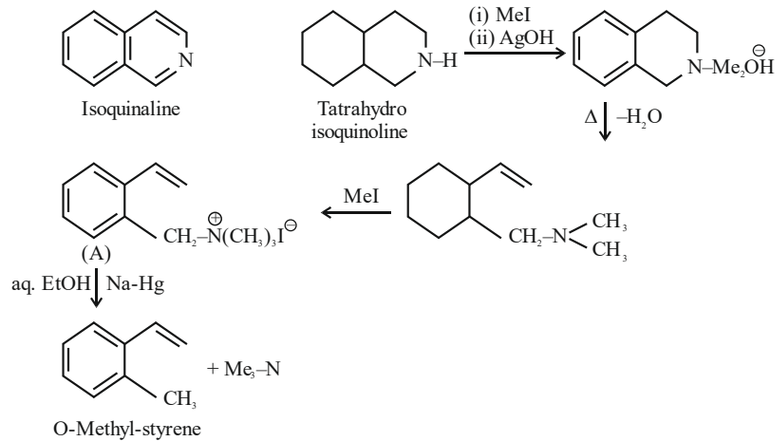


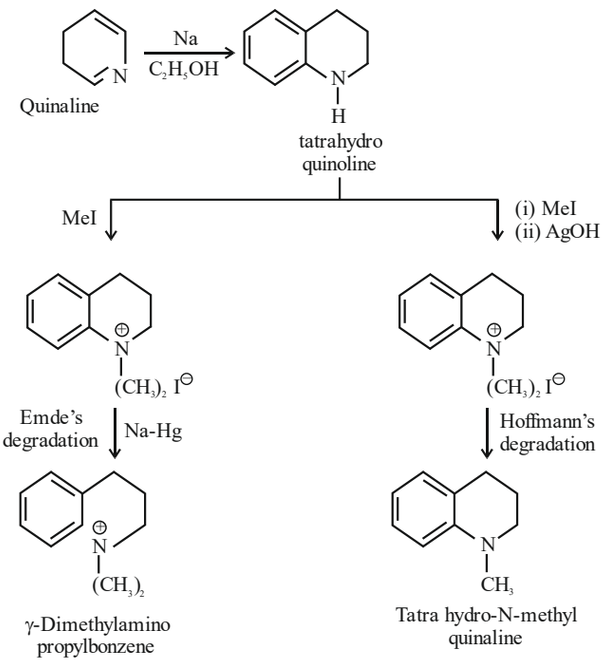
2. **Emde Degradation:** The Hoffmann's exhaustive methylation fails in cases where the alkaloids do not contain a β-hydrogen atom. In such cases, Emde degradation may be used successfully.

   In this degradation, alkaloid is converted to quaternary ammonium halide. The resulting halide is reductive cleavage by the treatment with sodium amalgam in alkanal or sodium in liquid ammonia or by catalytic hydrogenation.



   The intermediate 'A' cannot be degraded by Hoffmann's method as it has no β hydrogen but it can be degraded by Emde's method.

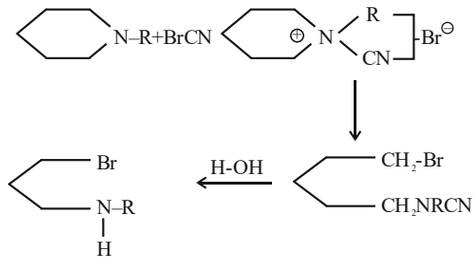
   In case of tetrahydroquinoline, though it contains a β-hydrogen, the Hoffmann's degradation fails.



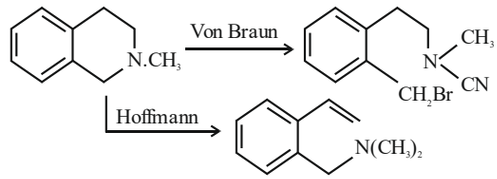
3. **Von Braun Degradation Method:** This method is used for those compounds which cannot be degraded by Hoffmann's degradation methods.

   This method involves the breaking of a heterocyclic ring. The heterocyclic rings containing the N atom is secondary and tertiary is nature.

   (i) **For Tertiary Nitrogen Atom:** When the tertiary nitrogen compound react with cyanogen bromide, cyanogen group is attached to the nitrogen atom and bromine atom is attached to carbon atom by the heating process brominated cyanamide derivative is formed and the process of hydrolysis it convert into brominated secondary amine.



   Von Braun's method cleave heterocyclic ring at different points:



   (ii) **The Von Braun Method for Secondary Cyclic Amine:** Secondary amine is treated with benzoyl chloride in presence of sodium hydroxide to form reaction mixture, it is treated with phosphorous halide to give

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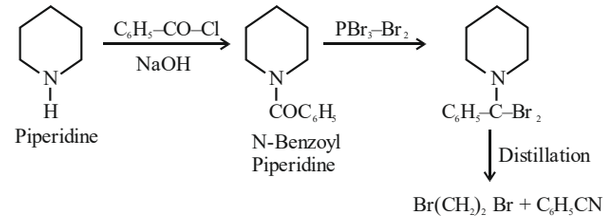
a dibromo derivative under the distillation process at reduced pressure it gives phenyl cyanide and dibromo alkane.



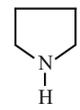
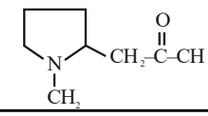
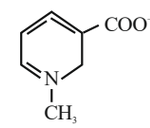
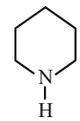
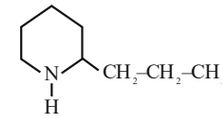
Check Your Progress

8. When the Hoffmann's exhaustive methylation fails?
9. Which method is used for those compounds which cannot be degraded by Hoffmann's degradation methods?
10. What is the nature of heterocyclic rings containing the N atom?

2.5 CLASSIFICATION BASED ON NITROGEN HETEROCYCLIC RING

1. On the basis of chemical structure: Alkaloid are divided into two groups:
   (A) **Atypical Alkaloids:** These are also known as non-heterocyclic alkaloids and contain nitrogen in aliphatic chain.
   Example: β-phenyl ethyl amine, benzedrine.
   (B) **Typical Alkaloids:** These are also known as the heterocyclic alkaloids and contain nitrogen in heterocyclic ring system. They are divided into two sub groups:
      (i) Mono-nuclear heterocyclic alkaloids
      (ii) Poly-nuclear heterocyclic alkaloids

Classification on the basis of nitrogen containing heterocyclic ring:

Name of Subgroups	Base Unit	Examples	Structure
Pyorolidine		Hygrine	
Pyridines piperidine	 Pyridine	Trigonelline	
	 Piperidine	Conine	

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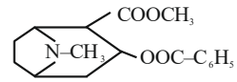
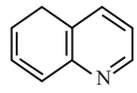
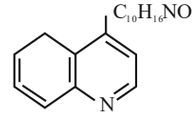
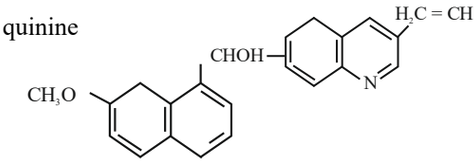
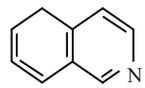
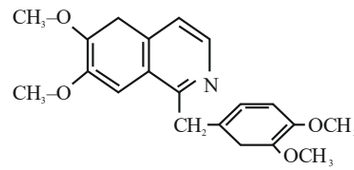
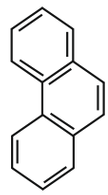
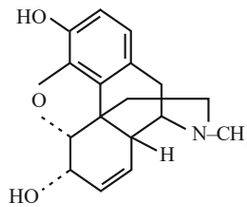
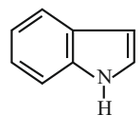
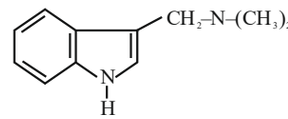
Tropane		Cacaine	
Quinocine		Cinchonine	
		quinine	
Isoquinaline		Papaverine	
Phenonthrene		Morphine	
Indole		Gramine	

2.5.1 Some Important Alkaloids

Here we will discuss some important alkaloids.

(A) Coniine (α-*n*-Propylpiperidine) [$C_8H_{17}N$]

Coniine is one of the simplest alkaloid known and was the first to be synthesized by Ladenburg in 1886. It occurs in seeds and other parts of the spotted hemlock (Conium maculatum) a shrub growing in wet places. Hemlock is a deadly poison. In 399 B.C. the great Greek philosopher, Socrates, was forced to meet his death by taking a cup of its extract. Seasonal variations seem to affect the coniine content, *e.g.*, green seeds contain 1.62%, half ripe 1.26% while ripe seeds upto 100% of total alkaloids. It occurs in the form of its salts with malic and caffeic acids.

Isolation. The crushed seeds of hemlock are distilled with sodium hydroxide. The distillate is extracted with other. The ether extract is concentrated and acidified with excess of acetic acid. The aqueous layer is again extracted with ether and basified with sodium hydroxide and re-extracted with ether. The ethereal extract is evaporated and the residue is carefully fractionated. The fraction distilling at 166°C is coniine.

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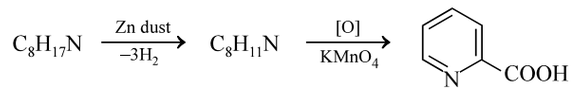
Properties. It is one of the few alkaloids, which are liquid (b.p. 166–167°C). It has unpleasant smell and gradually turns brown on exposure to air. It is insoluble in water, but fairly soluble in organic solvents. The natural product is dextrorotatory, $[\alpha]_D = +15.7°$. It is strongly basic and forms salts with acids like hydrochloric, hydrobromic etc.

Coniine as well as its salts are extremely poisonous. Death is caused because of the paralysis of the motor nerve endings, depression of central nervous system, an failure of respiratory system.

Constitution

1. From analysis and molecular weight determination, the molecular formula of coniine is found to be $C_8H_{17}N$.
2. When distilled with zinc dust, it gets converted to conyrine ($C_8H_{11}N$); the latter on oxidation with potassium permanganate forms pyridine-2-carboxylic acid (picolinic acid).



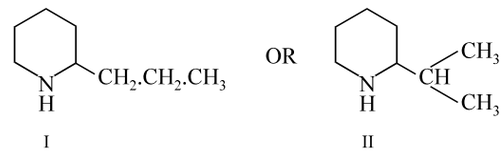
This indicates that:

(*i*) Conyrine contains a pyridine ring with a side-chain at position 2.

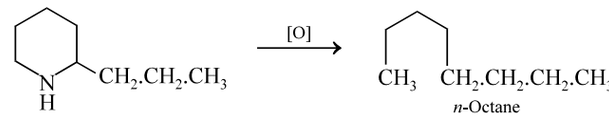
(*ii*) Conyrine contains six hydrogen atoms less than coniine, so the latter may probably be a piperidine derivative with a side-chain at 2-position.

(*iii*) Because coniine and piperidine have the formula $C_8H_{17}N$ and $C_5H_{10}N$ respectively, the side-chain may be —C_3H_7.

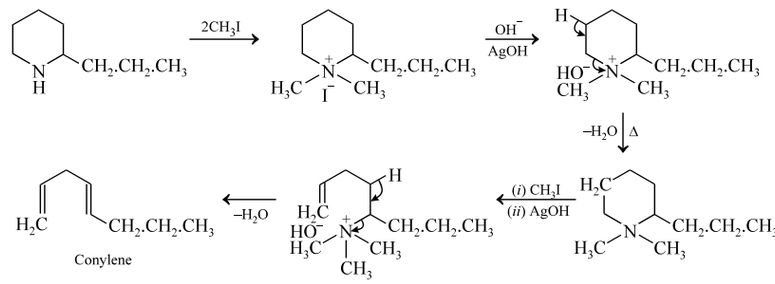
Thus coniine may be represented as:



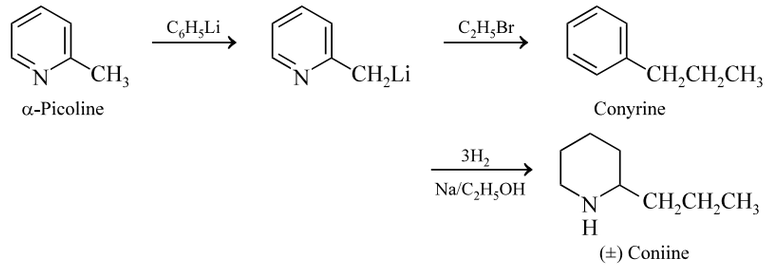
3. When heated with hydriodic acid at 300°C under pressure, coniine forms *n*-octane instead of iso-octane. This rules out the possibility of formula II.



4. Formation of conylene (C_8H_{14}) by exhaustive methylation (Hofmann degradation) further supports structure I.



5. The structure of coniine is finally confirmed by its synthesis, which is given below :



The racemic form (±) obtained above is resolved by means of (+)-tartaric acid and (+) -coniine thus obtained is found to be identical with the natural product.

(B) Nicotine (1-Methyl-2-β-pyridylpyrrolidine), [$C_{10}H_{14}N_2$]

As salt of malic and citric acids it occurs in tobacco plant, Nicotiana tabacu, and other Nicotiana species. In leaves of tobacco its concentration is the highest and varies from 0.6 to 8 per cent depending on the variety of tobacco. As a rule, the better kinds of tobacco contain smaller amount of nicotine.

Isolation: Tobacco leaves of high nicotine content are crushed and extracted with cold water. The hydrocarbons present in the extract are removed by acidifying the solution and extracting with ether. The residual solution is then made alkaline and the free nicotine is extracted with ether.

Properties: When freshly prepared, it is a colourless oily liquid (b.p. 246.2°C/ 730 mm). On exposure to air it rapidly turns brown due to oxidation. It has tobacco-like odour and a burning alkaline taste. It is soluble in water, as well as in organic solvents. The natural product is laevorotatory, $[\alpha]_D = -168.4°$. Aqueous solutions of its salts are, however dextrorotatory.

Nicotine is a deadly poison. Even two or three drops, taken internally, may cause death in a few minutes. In minute amount it stimulates the nervous system for a while, but after that there is too much of depression. It paralyses the nervous system, including the respiratory centre.

Its dilute solution is used as an insecticide for killing green fly, plant lice and mites on sheep and poultry.

Constitution

1. From analysis and molecular weight determination its molecular formula is found to be $C_{10}H_{14}N_2$.
2. With hydrochloric acid it forms the crystalline salt, nicotine dihydrochloride indicating that it is a diacid base.
3. On treatment with methyl iodide it forms dimethiodide, but does not form an acetyl or benzoyl derivative. This suggests that nicotine is a di-tertiary base.
4. Heating with hydriodic acid at 200–300°C and estimating methyl iodide formed as silver iodide, it is found to contain one —NCH_3 group (Herzig and Mayer method).

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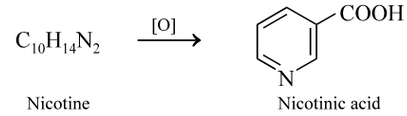
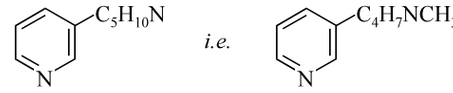
5. On oxidation with chromic acid or potassium permanganate, it forms nicotinic acid (pyridine-β-carboxylic acid). This suggests that nicotine is a pyridine derivative with a side-chain at β-position.

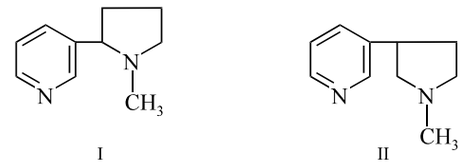


Nicotine Nicotinic acid

Hence nicotine may have the structure.

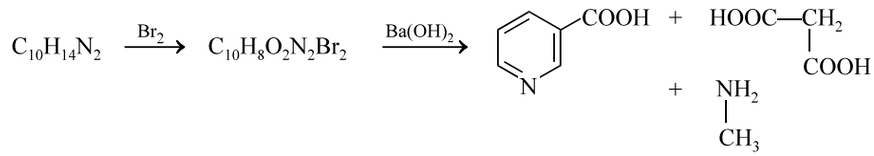


6. When nicotine-zinc chloride complex is distilled, the products are pyridine, pyrrole and methylamine. This suggests that the side-chain ($C_5H_{10}N$) should be a pyrrole derivative.
7. When reduced, nicotine takes up only six hydrogen atoms. As the six hydrogen atoms are utilized in converting pyridine residue to piperidine, the side chain —$C_4H_8NCH_3$ should be saturated and consequently cyclic. Accordingly nicotine may be assigned either of the two following structures.

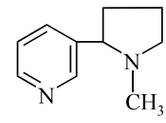


I II

8. Bromine in presence of hydrobromic acid converts nicotine into dibromonicotine, $C_{10}H_8O_2N_2Br_2$, which when heated with barium hydroxide at 100°C, yields nicotinic acid, malonic acid and methylamine.

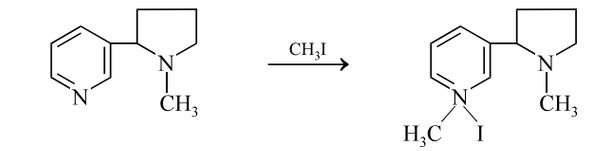


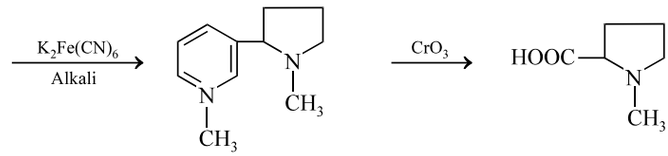
The formation of malonic acid, an acid with three carbon atoms, indicates that the carbon atom appearing as —COOH group in nicotinic acid must be the end atom of a chain of four carbon atoms. This is possible only when N-methylpyrrolidine moiety is linked to pyridine through α-carbon atom, hence it goes in favour of structure I.



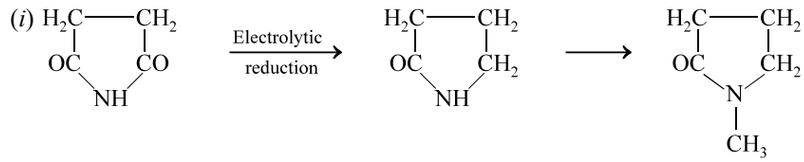
Nicotine

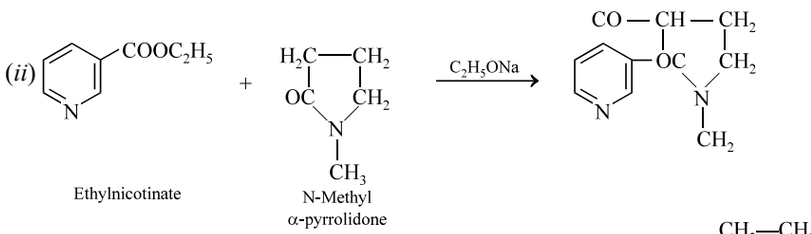
9. The above structure is further supported by the conversion of nicotine into *l*-hygrinic acid.

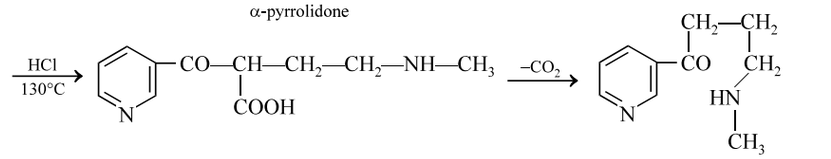


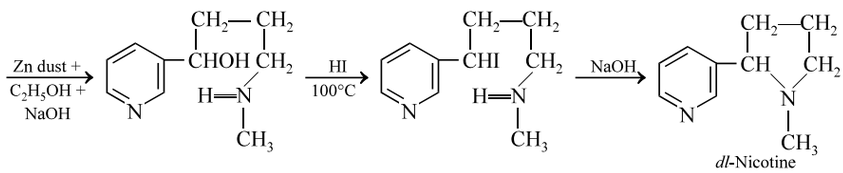


10. The structure was further confirmed by its synthesis by Spath and Bretschneider (1928).









On resolving the racemic nicotinic acid with *d*-tartaric acid, *l*-nicotine obtained as found to be identical with the natural product.

(C) Piperine, $C_{17}H_{19}O_3N$

Piperine was first prepared by Oersted in 1819. It occurs to the extent of 5 to 9 per cent in unripe fruit (black pepper) and in the kernel of ripe fruit (white pepper) of Piper nigrum. It is present in relatively smaller amount in other Piper species such as Piper longum (app. 5%), Piper lowong (app. 1.5%), etc.

Extraction. Dried black pepper is powdered and warmed with milk of lime for about 15 minutes. After evaporating the mixture to dryness on a water bath, the residue is extracted with ether. The ethereal solution is washed successively with

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sodium hydroxide and with water. The remaining ethereal solution is evaporated and the residue is crystallized from alochol to give pure piperine.

Properties. Piperine forms beautiful monoclinic colourless crystals (m.p. 128 – 129.5°C) having flavour of black pepper. It is practically insoluble in water, but soluble in the common organic solvents. It is itself tasteless and, therefore, not responsible for the particular taste of pepper. Actually an isomeric alkaloid, chavicine, occurring in pepper is responsible for the taste. It is practically neutral to litmus and is optically inactive. It is much less toxic than other alkaloids.

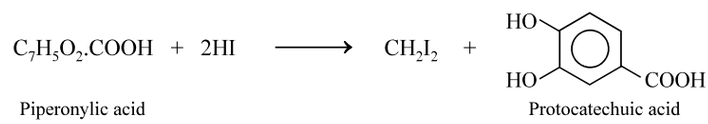
With acid or alkali it gets hydrolysed to piperic acid and piperidine.

Piperine is used as an antipyretic and for relieving colic pain.

Constitution

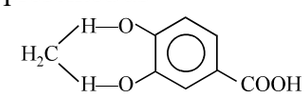
1. Analytical data and molecular weight determination indicate that it has the molecular formula $C_{17}H_{19}O_3N$.
2. When boiled with alcoholic alkali, piperine gets hydrolysed into piperic acid and piperidine (a secondary amine).

   This suggests that piperine is piperidine amide of piperic acid. Thus, knowing the structures of piperidine and piperic acid, that of piperine can be known.
3. **Structure of Piperidine.** Piperidine is hexahydropyridine. Its structure is further confirmed by exhaustive methylation, when a conjugate diene, piperylene, is formed and nitrogen is eliminated as trimethylamine.
4. **Structure of Piperic Acid**
   (*i*) Molecular formula of piperic acid is $C_{12}H_{10}H_4$.
   (*ii*) With sodium bicarbonate it gives effervescene, hence, it contains carboxyl group.
   (*iii*) The formation of tetrahydro and tetrabromo-piperic acid indicates the presence of two olefinic double bonds.
   (*iv*) On oxidation with potassium permanganate, it forms piperonal ($C_8H_6O_3$), piperonylic acid ($C_8H_6O_4$) and oxalic acid.
   (*v*) Piperonylic acid, when boiled with concentrated hydroidic acid, forms methylene iodide and protocatechuic acid (3,4-dihydroxy benzoic acid).

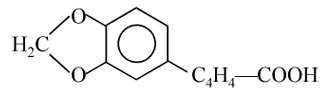


Piperonylic acid — Protocatechuic acid

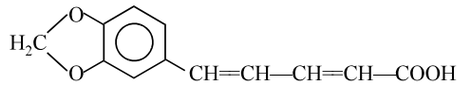
Hence, piperonylic acid is the methylene ether of protocatechuic acid and may be represented as



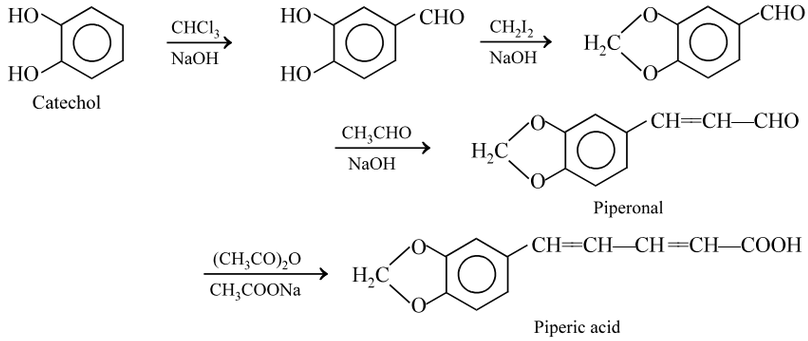
(*vi*) On the basis of facts (*iv*) and (*v*) piperic acid should, therefore, have the structure —



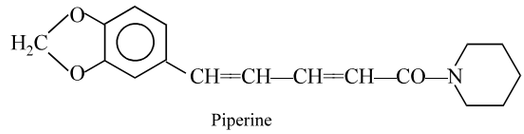
(*vii*) On ozonolysis piperic acid yields glyoxal and glyoxylic acid, therefore, the two double bonds in the side chain should be alternate, *i.e.*, the structure of piperic acid should be



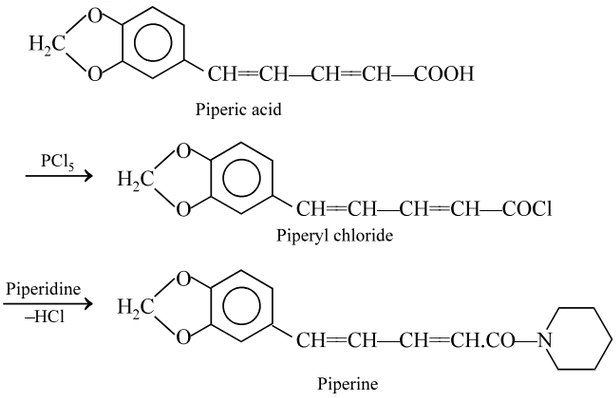
(*viii*) The structure of piperic acid is confirmed by its synthesis suggested by Ladenburg and Scholtz in 1894.



5. Thus, the structure of piperine, which is built up of piperic acid and piperidine units, linked through amide linkage, is



6. The structure of piperine is finally confirmed by its synthesis outlined below:



(D) Atropine, $C_{17}H_{23}O_3N$

Along with hyosycamine and other closely related alkaloids, atropine occurs in the berry of the deadly nightshade (Atropa belladona), in the thorn-apple (fruits of Datura stramonium), and in the henbane (Hyosycamus niger). These plants belong to Solanaceae family.

Extraction. Atropine is obtained from juice of deadly nightshade. The juice is mixed with potassium carbonate and extracted with chloroform. The latter is

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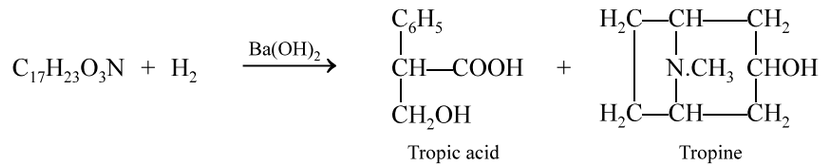
removed by evaporation. From the residue atropine is extracted with dilute sulphuric acid. The solution is then alkalified with potassium carbonate, when atropine gets precipitated. It is purified by recrystallization from alcohol.

Properties. Atropine has prismatic crystals (m.p. 115°C). It is insoluble in water, but readily soluble in alcoho, chloroform and ether. It is optically inactive.

It has a bitter taste and is extremely poisonous. It dilates the pupil of the eye, and is, therefore, extensively used in eye-surgery.

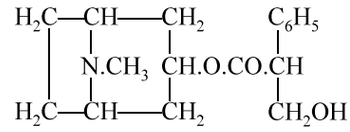
Constitution

1. From analysis and molecular weight determination, the molecular formula of atropine is found to be $C_{17}H_{23}O_3N$.
2. On hydrolysis with baryta-water it forms tropic acid and tropine.



Tropic acid Tropine

3. Hence, atropine is an ester of tropic acid and tropine and has the structure formula as :



Atropine

Test. Moisten a trace of atropine with fuming nitric acid in a porcelain dish. On evaporating it to dryness on a water-bath, a yellow stain is obtained. On adding a few drops of alcoholic caustic potash to the yellow stain, an intense violet colour develops, which gradually changes to red.

(E) Cocaine, $C_{17}H_{21}O_4N$

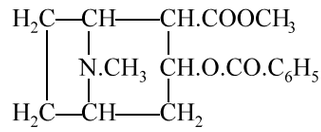
Cocaine occurs with several other closely related alkaloids in the leaves of Peruvian coca (Erythroxylon coca).

Extraction: The dried and powdered leaves of Peruvian coca are extracted with hot water at 80°C. Tannins, proteins, etc., are removed as precipitate with lead acetate. The liquid is filtered and lead is removed as lead sulphate, which is filtered off. The filtrate is alkalified and cocaine is extracted with ether and purified by crystallization with alcohol.

Properties: It forms colourless crystals (m.p. 98°C). It is laevorotatory and soluble in alcohol. It is a strong base and forms salts with acids. Its hydrochloride is readily soluble in water.

It is used in eye-surgery and dentistry as local anaesthetic.

Structural formula. Its structural formula is as given below.



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(F) Quinine, $C_{20}H_{26}O_2N_2$

Quinine, the well known anti-malarial drug occurs together with other alkaloids, *e.g.*, cinchonine, hydroquinine, cupreine, etc., in the bark of cinchona tree (Cinchona officinalis). The total alkaloid content of the bark is 6 per cent, out of which 70 per cent is quinine.

Quinine is a white solid, m.p. 177°C, which crystallises with three molecules of water (m.p. 57°C). It is almost insoluble in water but soluble in organic solvents. It has intensely bitter taste. The natural product is laevorotatory, $[\alpha]_D^{20°C} = -158.2°$.

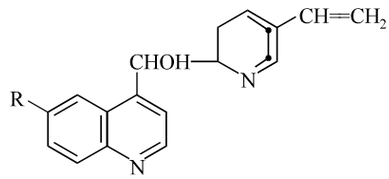
It is weak dibasic, but forms well-defined salts, such as, quinine sulphate, quinine hydrochloride, etc. The aqueous solution of these salts shows pale-blue fluorescence.

It is a potent anti-malarial drug and is also antipyretic.

Tests

1. Its dilute solution exhibits splendid blue fluorescence.
2. When a solution of quinine salt is treated with chlorine or bromine water and then ammonium hydroxide is added, a green precipitate is obtained, which soon dissolves in excess of ammonia forming an emerald-green colour.

Structure



R = OCH_3 : Quinine
R = H : Cinchonine

(G) Morphine, $C_{17}H_{19}O_3N$

It is the chief alkaloid present in opium obtained from poppy (Papaver somniferum).

It crystallizes from aqueous alcohol in colourless prisms, containing one molecule of water (m.p. 254°C). It is sparingly soluble in water and alcohol, but dissolves readily in alkalies, from which it may be reprecipitated by the addition of acids.

It possesses a bitter taste and is a monoacid base forming well crystallized salts.

It is used as a soporific and analgesic.

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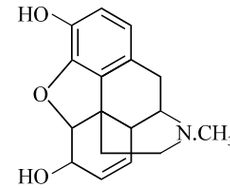
Tests

1. With ferric chloride morphine or its salts, it yields a deep blue colour.
2. If the solution of morphine is dissolved in concentrated sulphuric acid and is allowed to stand for 15 hours, and then some concentrated nitric acid is added, a deep bluish-violet colour develops, which gradually changes to blood-red.

Structure: Though almost the first crystalline alkaloid to be isolated by Serturner in 1805, its structure has been established only recently.



Morphine

(H) Strychnine, $C_{21}H_{32}O_2N_2$

The two highly poisonous alkaloids, strychnine and brucine are present in seeds of Strychnos nux vomica and Strychnos ignatii.

Strychnine is an indole alkaloid and crystallizes in beautiful colourless prisms, which decompose on heating at 245°C.

To its salt solution, when a little concentrated sulphuric acid and powdered potassium dichromate is added, an intense violet colour is produced, which gradually turns bright red and then yellow.

In minute amount it acts to stimulate the respiratory and vasomotor rerve centres. Being extremely poisonous it is used as pest exterminator.

Check Your Progress

11. What are atypical alkaloids?
12. Into which two sub groups' typical alkaloids are divided?
13. Name an alkaloid which is in liquid state.
14. Which alkaloid is present in the various species of pepper?

2.6 ROLE OF ALKALOIDS IN PLANTS

Alkaloids are one of the largest groups of plant secondary metabolites. These are present in several plant families. These are end products of metabolism and are waste products of the plant, it was considered that alkaloids have diverse physiological effects on humans, plants and other animals

1. Alkaloids can act as defence compounds in plant. These are efficient against pathogens.
2. Alkaloids play role in plant metabolism plant catabolism or in plant physiology these act as growth regulators.

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3. The presence of alkaloids in plants enhance plant reproductive rates by introducing defences against biotic and abiotic stresses by affecting pollinators and seed.
4. These leave stimulant properties.
5. The major role of alkaloids in plants is protection against herbivores. Alkaloids has bitter flavour, disruption of protein function after ingestion and metabolization
6. Alkaloids specific toxicity can be used to fight tumour cell.
7. In environments, such as, those with temperature, floods or droughts, mechanism to tolerate freezing and dormancy period to prevent loss of water and deal with anoxia.
8. Alkaloids has wide range of physiological effects on animals. It has antibiotic activity.
9. These are toxic to insects.
10. Nicotine from tobacco is one of the first insecticides and is most effective against herbivory.
11. Caffeine is also an effective insect toxin. These are mostly found in leaves and beans of cocoa and coffee.
12. Caffeine kills larvae of the tobacco hornworm within 24 hours in dietary concentration.
13. Alpha-solanine is a cholinesterase inhibitor.

Check Your Progress

15. How the presence of alkaloids in plants enhance plant reproductive rate?
16. Which alkaloid is found in leaves and beans of cocoa and coffee?
17. Nicotine is extracted from which plant?

2.7 ANSWERS TO 'CHECK YOUR PROGRESS'

1. The term alkaloid, meaning alkali like, usually applies to basic nitrogenous substances of plant origin, with marked physiological action.
2. Alkaloids occur chiefly in dicotyledonous plants as salts of common plant acids, such as, malic, citric, oxalic, succinic or tannic and sometimes of special acids like quinic acid in the alkaloid of cinchona bark and meconic acid in those of morphine group.
3. Alkaloids are generally colourless crystalline solids which can not be distilled. A few of them eg. nicotine and coniine are liquid and volatize without decomposition.
4. Alkaloids are strong base.
5. Presence of unsaturation or double bond is determined by addition reaction with Br_2, H_2 or with halos acids

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6. When alkaloid is heated with concentrated solution of HI at 126°C. The methoxy groups are convert into methyl iodide.
7. The presence of N-methyl group is often detected by distillation of amine with soda lime or estimated by the treatment with HCl at 150–300°C and conversion of methyl iodide produced to silver iodide as mentioned for estimation of methoxy group.
8. The Hoffmann's exhaustive methylation fails in cases where the alkaloids do not contain a β-hydrogen atom.
9. Von Braun degradation method is used for those compounds which cannot be degraded by Hoffmann's degradation methods.
10. The heterocyclic rings containing the N atom is secondary and tertiary is nature.
11. Atypical alkaloids are also known as non-heterocyclic alkaloids and contain nitrogen in aliphatic chain.
12. Typical alkaloids are divided into two sub groups:
    (i) Mono-nuclear heterocyclic alkaloids
    (ii) Poly-nuclear heterocyclic alkaloids
13. Coniine is one of few alkaloids which are in liquid state.
14. Piperine is present in the various species of pepper.
15. The presence of alkaloids in plants enhance plant reproductive rates by introducing defences against biotic and abiotic stresses by affecting pollinators and seed.
16. Caffeine is found in leaves and beans of cocoa and coffee.
17. Nicotine is extracted from tobacco plant.

2.8 SUMMARY

- The term alkaloid, meaning alkali like, usually applies to basic nitrogenous substances of plant origin, with marked physiological action.
- Structurally alkaloids have great diversity, so no convenient and systematic method is possible for naming them.
- Alkaloids are usually classified according to the nature of heterocyclic ring containing nitrogen atom.
- Alkaloids occur chiefly in dicotyledonous plants as salts of common plant acids such as malic, citric, oxalic, succinic or tannic and sometimes of special acids like quinic acid in the alkaloid of cinchona bark and meconic acid in those of morphine group.
- Knowing the presence of nitrogen and/or oxygen in the alkaloid, the nature of the functional group is determined.
- With qualitative and quantitative analysis empirical formula of the alkaloid compound is determined.

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- Primary alcoholic group on oxidation of alkaloid, gives an aldehyde and then acid with the same number of carbon atom as the original molecule.
- The Hoffmann's exhaustive methylation fails in cases where the alkaloids do not contain a β-hydrogen atom. In such cases, Emde's degradation may be used successfully.
- Coniine is one of the simplest alkaloid known and was the first to be synthesized by Ladenburg in 1886. It occurs in seeds and other parts of the spotted hemlock (Conium maculatum) a shrub growing in wet places.
- From analysis and molecular weight determination, the molecular formula of coniine is found to be $C_8H_{17}N$.
- Nicotine is a deadly poison. Even two or three drops, taken internally, may cause death in a few minutes.
- With hydrochloric acid nicotine forms the crystalline salt, nicotine dihydrochloride indicating that it is a diacid base.
- Piperine was first prepared by Oersted in 1819. It occurs to the extent of 5 to 9 per cent in unripe fruit (black pepper) and in the kernel of ripe fruit (white pepper) of Piper nigrum.
- Molecular formula of piperic acid is $C_{12}H_{10}H_4$.
- Quinine, the well-known anti-malarial drug occurs together with other alkaloids, e.g., cinchonine, hydroquinine, cupreine, etc., in the bark of cinchona tree (Cinchona officinalis).
- Alkaloids play role in plant metabolism plant catabolism or in plant physiology these act as growth regulatory.
- The presence of alkaloids in plants enhance plant reproductive rates by introducing defences against biotic and abiotic stresses by affecting pollinators and seed.

2.9 KEY TERMS

- **Alkaloid:** The term *alkaloid*, meaning *alkali like*, usually applies to basic nitrogenous substances of plant origin, with marked physiological action.
- **Atypical Alkaloids:** These are also known as non-heterocyclic alkaloids and contain nitrogen in aliphatic chain.
- **Typical Alkaloids:** These are also known as the heterocyclic alkaloids and contain nitrogen in heterocyclic ring system.
- **Coniine:** Coniine is one of the simplest alkaloid known and was the first to be synthesized by Ladenburg in 1886.
- **Quinine:** Quinine is a well-known anti-malarial drug occurs in the bark of cinchona tree (Cinchona officinalis).

2.10 SELF-ASSESSMENT QUESTIONS AND EXERCISES

NOTES

Short-Answer Questions

1. Where does alkaloids chiefly occur?
2. State about solubility of alkaloids.
3. How the presence of hydroxyl group is established in alkaloids?
4. What is Hofmann Exhaustive Methylation?
5. Write the reactions of Emde's degradation.
6. What are the properties of nicotine?
7. How piperine is extracted from black pepper?
8. What is the molecular formula of nicotine?

Long-Answer Questions

1. Explain the general characters of alkaloids.
2. Describe the general outline of the isolation of alkaloids from plant material.
3. Describe the steps used to establish the molecular structure of alkaloids.
4. Comprehend the classification of alkaloids on the basis of nitrogen containing heterocyclic ring.
5. Analyse the role of alkaloids in plants.

2.11 FURTHER READING

Krishnaswamy, N. R. 1999. *Chemistry of Natural Products A Unified Approach*. Himayatnagar, Hyderabad: Universities Press.

Bhat, Sujata V., B.A. Nagasampagi, Meenakshi Sivakumar · 2005. *Chemistry of Natural Products*. Narosa Publishing House

Rahman,Atta-ur. 2020. *Studies in Natural Products Chemistry.* Volume 67. Amsterdam, Netherlands: Elsevier Science.

Cooper, Raymond. , George Nicola. 2014. *Natural Products Chemistry-Sources, Separations and Structures.* London UK: Taylor & Francis

UNIT 3 STEROID

NOTES

Structure

3.0 INTRODUCTION

Steroid refers to any of a class of natural or synthetic organic compounds characterized by a molecular structure of 17 carbon atoms arranged in four rings. Steroids are important in biology, chemistry, and medicine. Hundreds of steroids are found in plants, animals and fungi. All steroids are manufactured in cells from the sterols lanosterol (opisthokonts) or cycloartenol (plants). Lanosterol and cycloartenol are derived from the cyclization of the triterpene squalene. The steroid group includes all the sex hormones, adrenal cortical hormones, bile acids, and sterols of vertebrates, as well as the molting hormones of insects and many other physiologically active substances of animals and plants. Among the synthetic steroids of therapeutic value are a large number of anti-inflammatory agents, anabolic (growth-stimulating) agents, and oral contraceptives. Different categories of steroids are frequently distinguished from each other by names that relate to their biological source, e.g., phytosterols (found in plants), adrenal steroids, and bile acids or to some important physiological function e.g., progesterone (promoting gestation), androgens (favouring development of masculine characteristics), and cardiotonic steroids (facilitating proper heart function).

In this unit, we will discuss the occurrence, nomenclature, and basic skeleton of steroids, along with the Diel's hydrocarbon and stereochemistry. It will also focus

on the isolation, structure determination, and synthesis of cholesterol, bile acids, androsterone, testosterone, estrone, progesterone, and aldosterone.

NOTES

3.1 OBJECTIVES

After going through this unit, you will be able to:

- Describe the occurrence, nomenclature, and basic skeleton of steroids
- Explain the isolation, structure determination, and synthesis of cholesterol, bile acids, androsterone, and testosterone
- Discuss the isolation, structure determination, and synthesis of estrone, progesterone, and aldosterone

3.2 CONCEPT OF STEROIDS

Any of various compounds containing a 17-carbon 4-ring system and including the sterols and numerous hormones (such as, anabolic steroids or corticosteroids) and glycosides are steroids. A steroid is a biologically active organic compound with four rings arranged in a specific molecular configuration. Any of a large group of fat-soluble organic compounds, as the sterols, bile acids, and sex hormones, most of which have specific physiological action are considered as steroids

Steroids vary from one another in the nature of attached groups, the position of the groups, and the configuration of the steroid nucleus (or gonane). Small modifications in the molecular structures of steroids can produce remarkable differences in their biological activities. A steroid is a biologically active organic compound with four rings arranged in a specific molecular configuration. Steroids have two principal biological functions: as important components of cell membranes which alter membrane fluidity; and as signaling molecules. Hundreds of steroids are found in plants, animals and fungi. All steroids are manufactured in cells from the sterols lanosterol (opisthokonts) or cycloartenol. Lanosterol and cycloartenol are derived from the cyclization of the triterpene squalene.

3.2.1 Occurrence

Natural steroids are compounds that mimic the steroids that human bodies naturally produce, such as, the hormones testosterone, progesterone, etc. They are used to build muscle tissue and repair muscle by increasing the production of testosterone, among other important bodily processes. Natural steroids may also be called legal steroids, and businesses often sell them as a mixture of ingredients. Compounds with some preliminary evidence to support them include the following. The steroid's core structure is typically composed of 17 carbon atoms arranged in four fused rings. Consist of three fused six-membered and one five membered ring.

Steroids vary by the functional groups attached to this four-ring core and by the oxidation state of the rings. Sterols are forms of steroids with a hydroxy group at position three and a skeleton derived from cholestane. Steroids can also be more radically modified, such as by changes to the ring structure, for example, cutting one of the rings. Cutting ring B produces secosteroids one of which is vitamin

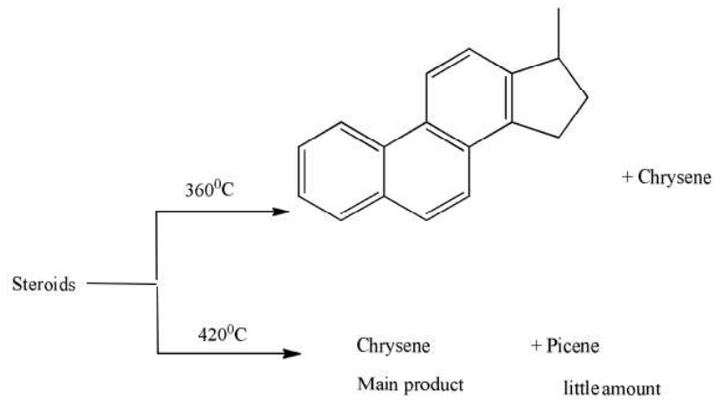
D3. Examples include the lipid cholesterol, the sex hormones estradiol and testosterone, and the anti-inflammatory drug dexamethasone.

A steroid is a biologically active organic compound with four rings arranged in a specific molecular configuration. Steroids have two principal biological functions: as important components of cell membranes which alter membrane fluidity; and as signalling molecules. Hundreds of steroids are found in plants, animals and fungi. All steroids are manufactured in cells from the sterols lanosterol or cycloartenol (plants). Lanosterol and cycloartenol are derived from the cyclization of the triterpene squalene.

NOTES

Steroids are compounds most of them isolated from animal sources and some of them isolated from plant source. Steroids produces Diels hydrocarbon and small amount of chrysene on distillation with Selenium at 360°C. If distillation carried out at 420°C, chrysene is the main product with small amount of picene. All steroids containing per hydro cyclo penteno phenanthrene with molecular formula CnH_2n-6. Steroids include:

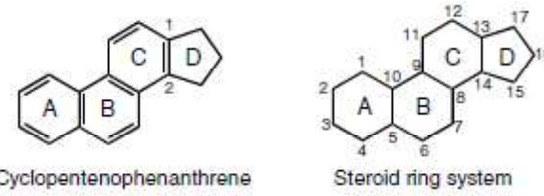
- Sterols
- Steroid hormones
- Vitamins
- Bile acids
- Adrenal cortex hormones
- Some carcinogenic hydrocarbon



3.2.2 Basic Skeleton of Steroid

The steroids are a family of compounds widely distributed in plants and animals. Common to the structure of all compounds of this class is a tetracyclic framework composed of the phenanthrene nucleus to which is fused at the 1, 2-positions a cyclopentene ring. The rings in the steroid molecule usually are not aromatic but often contains one or more isolated double bonds. The total structure of one steroid differs from that of another, usually by a variation in the side chain or by a variation in the number and type of functional groups. To the family of steroids with this common ring system belong the sterols, the sex hormones, the bile acids, and other biologically important materials. For purposes of nomenclature the steroid ring skeleton is numbered as given below.

NOTES



The steroid core structure is typically composed of seventeen carbon atoms, bonded in four "fused" rings: three six-member cyclohexane rings (rings A, B and C in the first illustration) and one five-member cyclopentane ring (the D ring). Steroids vary by the functional groups attached to this four-ring core and by the oxidation state of the rings. Sterols are forms of steroids with a hydroxy group at position three and a skeleton derived from cholestane. Steroids can also be more radically modified, such as by changes to the ring structure, for example, cutting one of the rings. Cutting Ring B produces secosteroids one of which is vitamin D3.

3.2.3 Steroid Nomenclature

Gonane, also known as steran or cyclopentanoperhydrophenanthrene, the simplest steroid and the nucleus of all steroids and sterols, is composed of seventeen carbon atoms in carbon-carbon bonds forming four fused rings in a three-dimensional shape. The three cyclohexane rings (A, B, and C in the illustration) form the skeleton of a perhydro derivative of phenanthrene. The D ring has a cyclopentane structure. When the two methyl groups and eight carbon side chains (at C-17, as shown for cholesterol) are present, the steroid is said to have a cholestane framework. The two common 5α and 5β stereoisomeric forms of steroids exist because of differences in the side of the largely planar ring system where the hydrogen (H) atom at carbon-5 is attached, which results in a change in steroid A-ring conformation. Isomerisation at the C-21 side chain produces a parallel series of compounds, referred to as isosteroids.

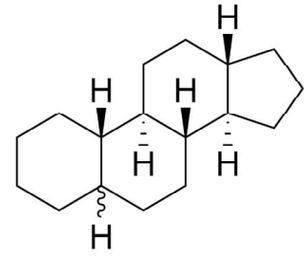
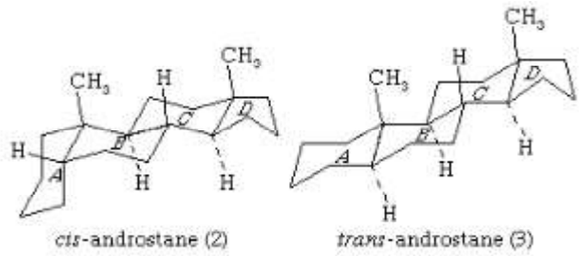
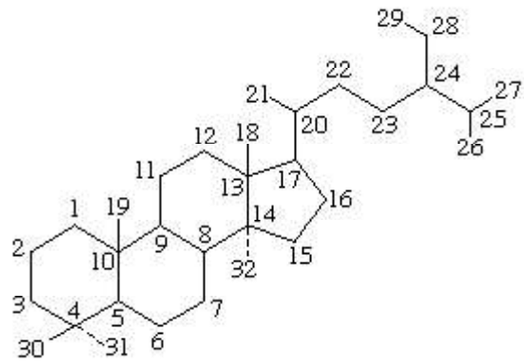


Fig 3.1 *Gonane, the Simplest Steroid, Consisting only of the Common Steroid Nucleus*

The steroid nucleus is a three-dimensional structure, and atoms or groups are attached to it by spatially directed bonds. Although many stereoisomers of this nucleus are possible (and may be synthesized), the saturated nuclear structures of most classes of natural steroids are alike, except at the junction of rings A and B. For example, androstane, common to a number of natural and synthetic steroids, exists in two forms (2 and 3), in which the A/B ring fusions are called cis and trans, respectively.



The stereochemistry of rings A and B must be specified by showing the orientation of the hydrogen atom attached at C5 (that is, carbon atom number 5; steroid numbering is explained below) as either above the plane of the diagram (designated β) or below it (α). The α- β- symbolism is used in a similar manner to indicate the orientation of any substituent group that is attached to a saturated (fully substituted) carbon within the steroid ring system. Groups attached to unsaturated carbons lie in the same plane as the adjacent carbons of the ring system (as in ethylene), and no orientation need be specified. When the orientation of a substituent is unknown, it is assigned the symbol ξ. Each carbon atom of a steroid molecule is numbered, and the number is reserved to a particular position in the hypothetical parent skeletal structure (6) whether this position is occupied by a carbon atom or not.



3.2.4 Types of Steroids

Steroids are classified into mainly three heads which are plant steroids, animal steroids, and fungal steroids. Plant steroids are the steroids that help in the growth and maintenance of the plant structure. It includes various alkaloids which are found in Melanthiaceae and Solanaceae. Other plant hormones that contain steroids in the form of alkaloids are brassino steroids and phytosterols. Animal steroids are those compounds which are mostly produced by the animal body. A common example of animal steroids is the sex hormone, such as, testosterone, estrogen, progestogen, etc. which helps in reproduction and sex determination. Another example of animal steroid is anabolic steroids. Anabolic steroids are those steroids that help in muscle and bone functioning and development. Fungal steroids are used to kill micro-organisms such as bacteria, fungi, and other pathogens. Some fungal steroids which are produced inside fungi help microorganism to grow and develop an ability to alter the effect of those steroids which are used to kill them. The stereochemistry of all these steroids differs due to the difference in the orientation of groups.

NOTES

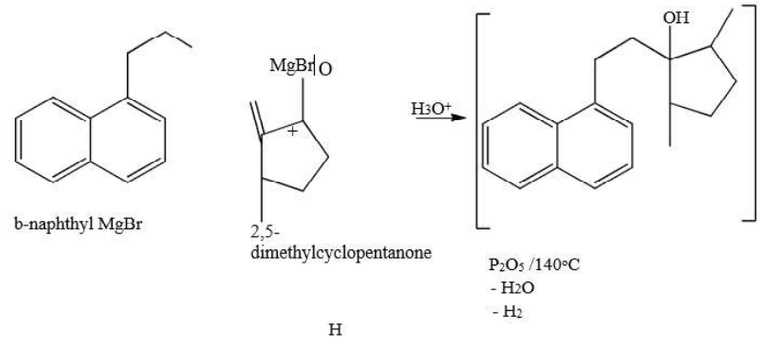
3.2.5 Stereochemistry of Steroids

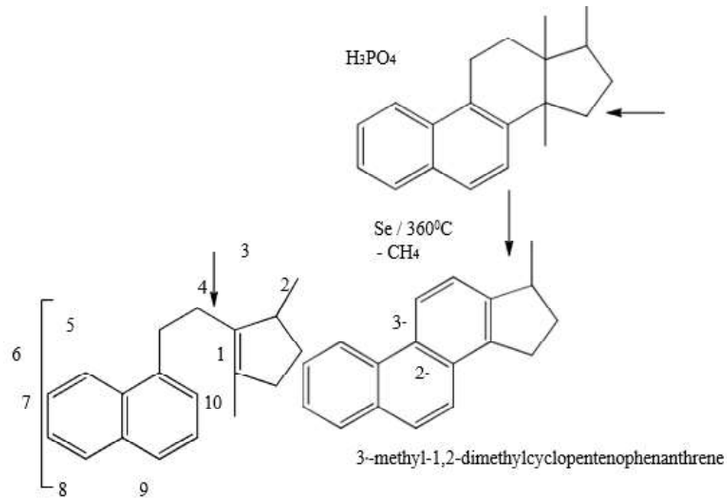
Stereochemistry of the steroids can be defined as the spatial arrangement of the substituents of steroids. In biological processes, the molecules which are responsible for such processes are chiral compounds. The activity and the acceptance of the compound by the body is purely based on the stereochemistry of the biomolecule. Steroids are one of those biomolecules which are required by the body and their stereochemistry plays a vital role in their binding and functioning in the living bodies. Cholesterol molecule have eight chiral carbon atoms. As the functioning of the biomolecule depends upon the stereochemistry, they are semi synthetically modified in the laboratories to develop drugs which are accepted by the body for proper functioning. The IUPAC name of cholesterol is (3S,8S,9S,10R,13R,14S,17R)-10,13-dimethyl-17-[(2R)-6-methylheptan-2-yl]-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1H-cyclopenta phenanthren-3-ol. The IUPAC name of testosterone is (8R,9S,10R,13S,14S,17S)-17-Hydroxy-10,13-dimethyl-1, 2, 6, 7, 8, 9, 11, 12, 14,15,16,17-dodecahydrocyclopenta[a]phenanthren-3-one.

Determination of Stereochemistry of Steroids

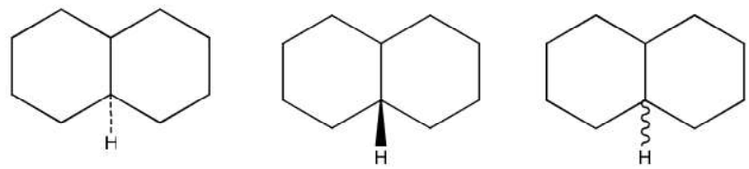
The basic method of determination of the stereochemistry is to find the optical activity in a molecule. If any molecule is optically active, then further determination processes can be employed. Chiral carbon atoms must be determined which provides clear information about the stereochemistry of the molecule. Based on the chiral carbon atoms, two methods can be followed. One is analytical and the other one is digital- instrumental. In an analytical method, the projection of the molecule is determined, and the molecule is drawn in a paper in either Fischer projection or Haworth projection. By following this method, the chirality of the molecule and hence the stereochemical aspect of the molecule can be understood. The other method which is more commonly used now a days is the instrumental method. A direct technique which is called chiral mass spectrometry is employed for the determination of the stereochemistry of the molecule. The other techniques which may add to the results obtained from chiral mass spectrometry are IR and NMR spectroscopy based on the interaction of the substituents with each other. Intramolecular hydrogen bonding can be reflected in both the IR and NMR spectrum of the molecule. In steroids, it is observed that the effect of the interaction of the substituents is detected in the analyzing techniques

3.2.6 Diels hydrocarbon and Stereochemistry (3'-methyl-1, 2 cyclopentenophenanthrene





Some nomenclature of steroids:



For substituents:

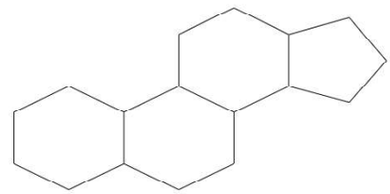
Dotted line refers to α-configuration; Solid line refers to β-configuration; Wavy line refers to unknown configuration.

Sterols

They are present in animal and plant in oils and fats and are of three types:

- Zoosterols: They are sterols of animals ,
- Phytosterols: They are sterols of plants
- Mycosterols: They are sterols of yeast and fungi

All sterols are containing Diel's hydrocarbon nucleus or per hydro cyclo penteno phenanthrene with molecular formula $C_nH_{2n-2} = C_{17}H_{28}$



1-Cholesterol $C_{27}H_{46}O$

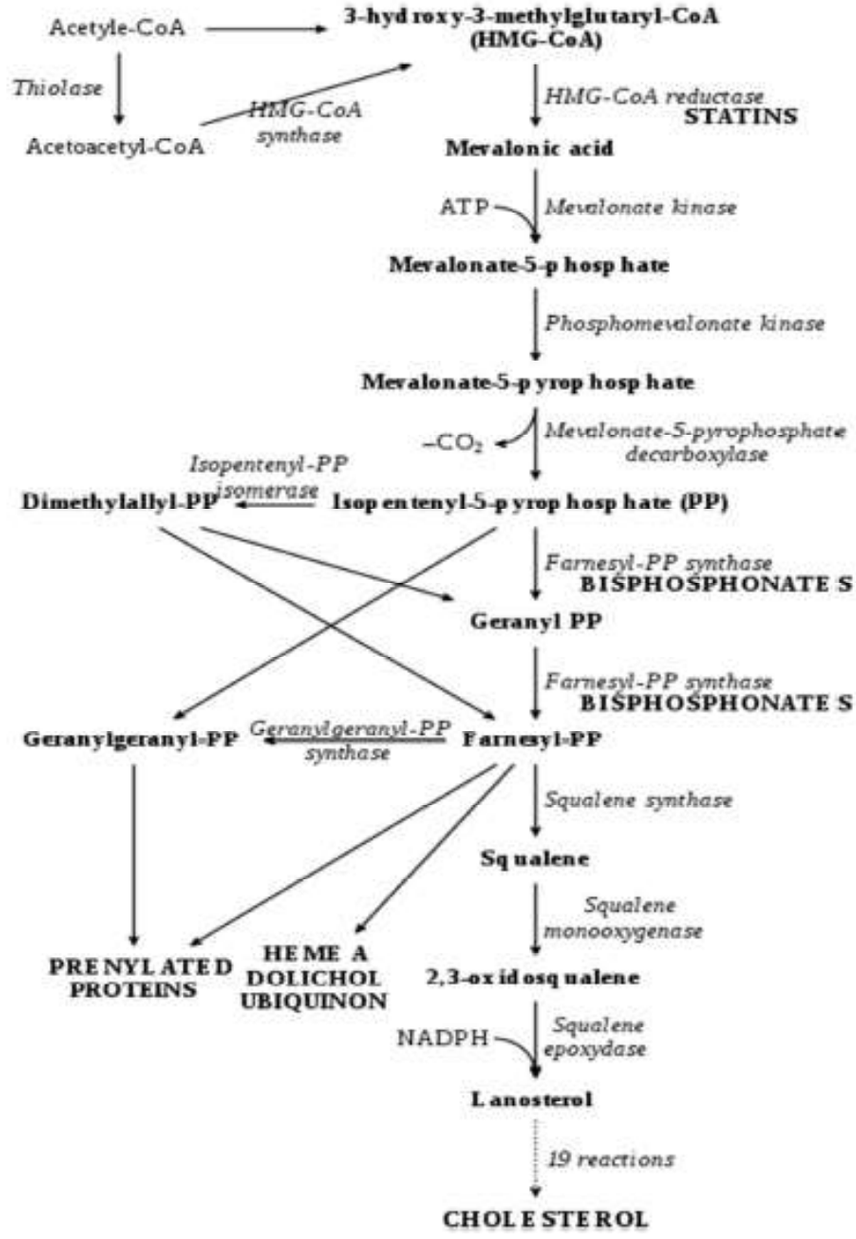
3.2.7 Biosynthesis of Steroids

This process occurs in multi-step and is catalyzed by enzyme. In this process substrate converted into complex products in living organism. In biosynthesis simple compounds are converted into other compounds and joined together to form macromolecules like production of lipid membrane and nucleotides it's also called Anabolism. Biosynthesis required substrate, chemical energy, Catalytic enzyme, Coenzyme or Cofactors, such as, NADH.

NOTES

Steroids contains a class of sterols, cholesterol is one of them. Biosynthesis of cholesterols has three stages, first stage takes place in cytoplasm and the second and third stages occurs in endoplasmic reticulum.

- The synthesis of the building block of cholesterol which is isopentenyl pyrophosphate.
- Isopentyl phosphate gets condensed and form squalene.
- After many enzymatic reactions squalene converts into cholesterol.



Check Your Progress

1. What are the principal biological functions of steroids?
2. Define stereochemistry of the steroids.

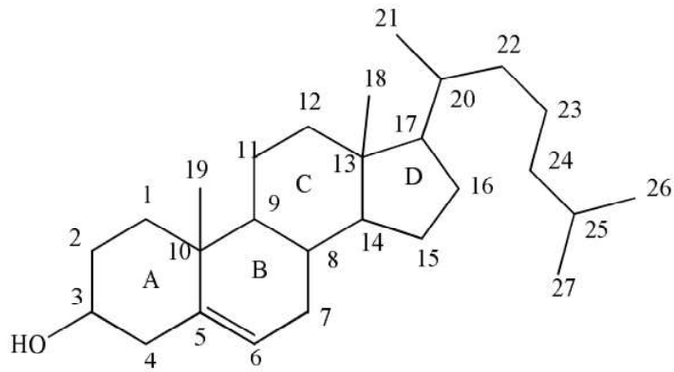
3.3 ISOLATION, STRUCTURE DETERMINATION AND SYNTHESIS OF DIFFERENT STEROIDS

NOTES

In this section we will discuss the isolation, structure determination, and synthesis of some of the common steroids.

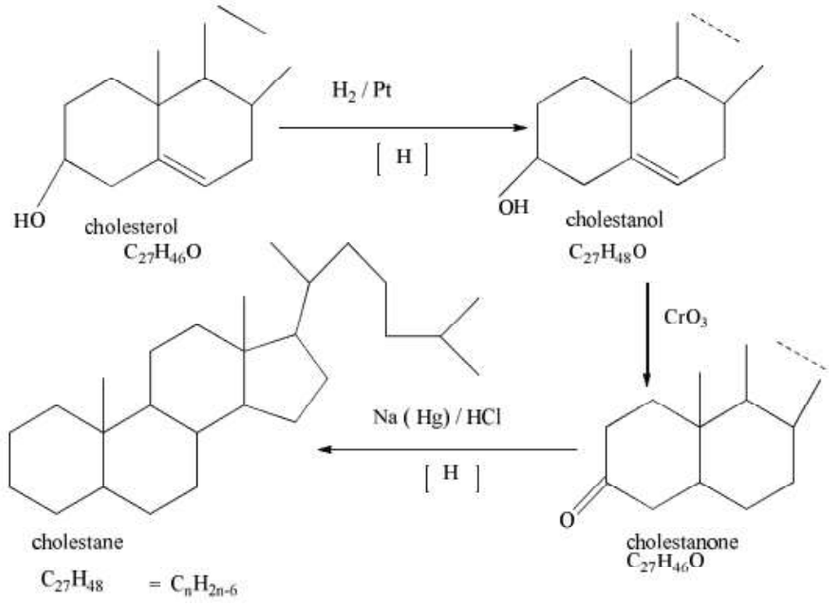
3.3.1 Cholesterol ($C_{27}H_{46}O$)

Cholesterol refers to a group of organic molecules. A cholesterol is a form of lipid known as a sterol (or modified steroid). All animal cells produce cholesterol, which is an important structural component of cell membranes. It is a yellowish crystalline solid when chemically separated.



It is present in the animal cell free or as fatty esters especially in the brain and spinal cord. Colour reactions of cholesterol gives a solution of cholesterol in chloroform gives a red colour with concentrated H_2SO_4 and gives greenish colour with concentrated H_2SO_4 / $(CH_3CO)_2O$.

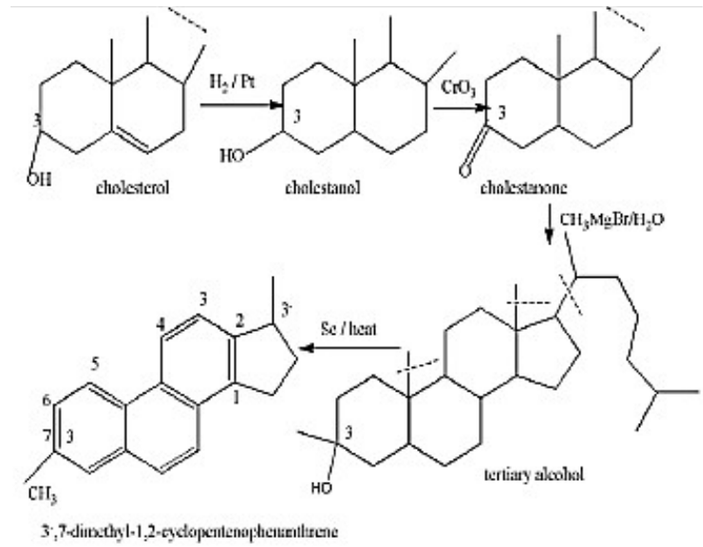
Structure of the Ring System (Tetracyclic Form of Cholesterol)



NOTES

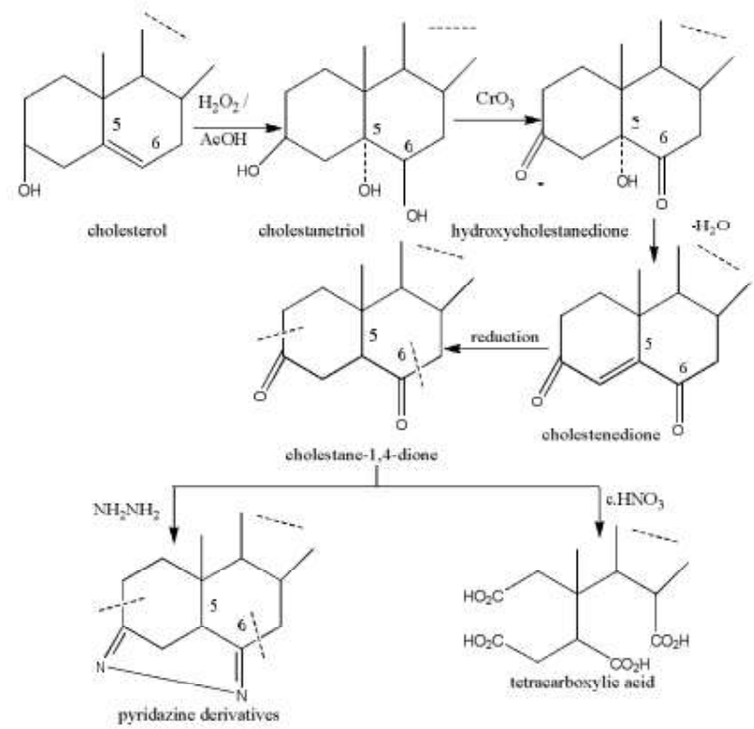
Formation of cholestanol from cholesterol indicates the presence of one double bond in cholesterol. Reduction of cholestanone to cholestane indicates the presence of a secondary hydroxyl group in cholesterol. Consequently, cholesterol is a tetracyclic ring compound with one double bond and secondary hydroxyl group. Distillation of cholesterol with selenium give Diels hydrocarbon, indicates the presence of this nucleus in it.

- Position of the hydroxyl group (at C-3)



Thus, the hydroxyl group in cholesterol at C-3.

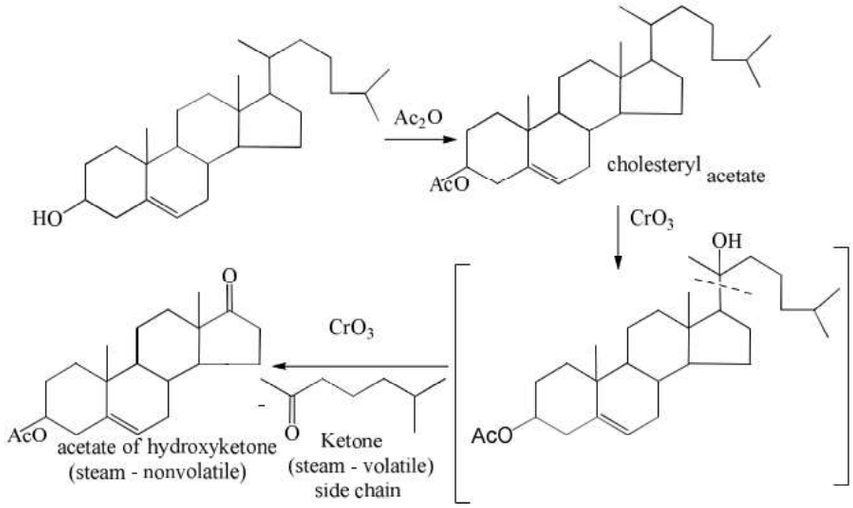
- Position of the double bond can be indicated by the following reactions:



Consequently, the double bond at C_5—C_6 in the ring.

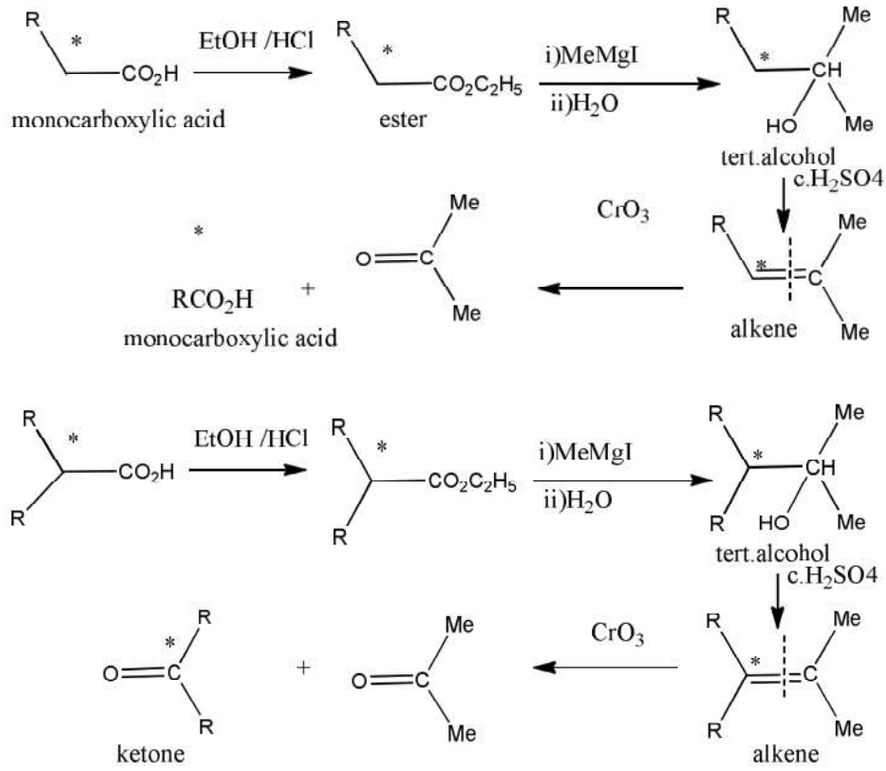
NOTES

- The nature and position of the side chain in cholesterol it has been found that:



The side chain is methylisohexylketone (C8) and the nucleus is (C19)

Barbier-Wieland Degradation (B.W.)



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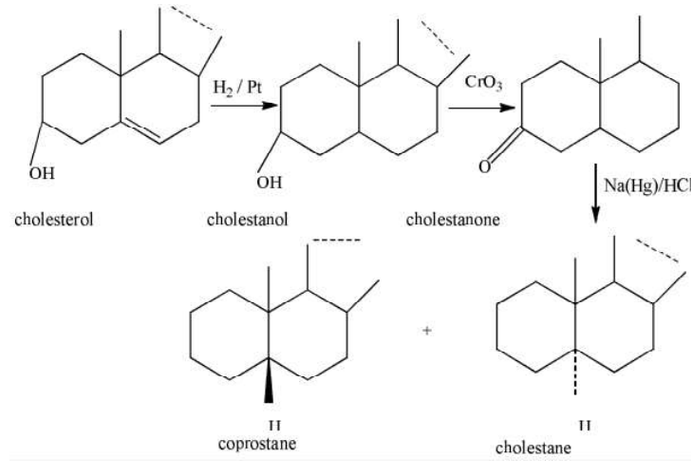
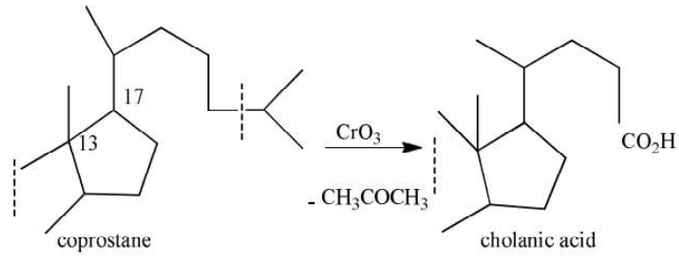
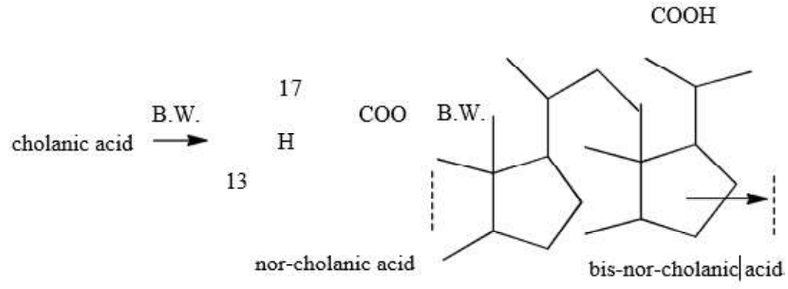
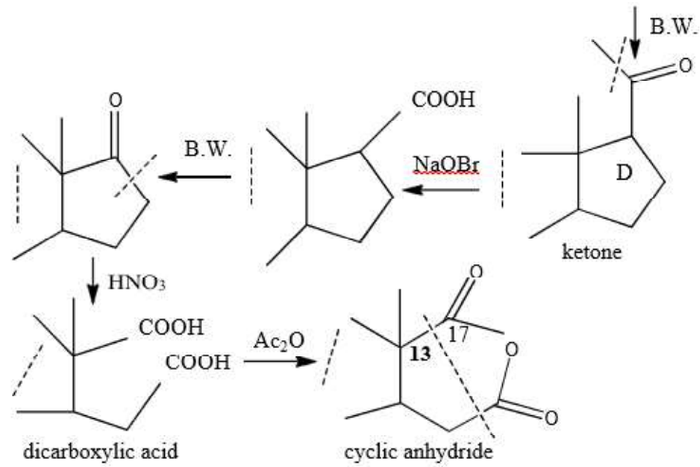


Degradative oxidation for the side chain in coprostane



Formation of acetone means that the side chain of cholesterol ends with isopropyl group.





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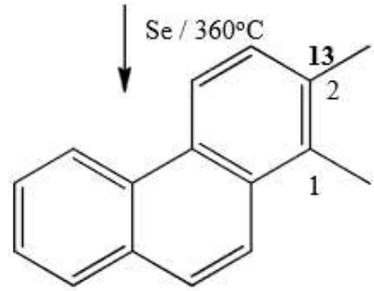
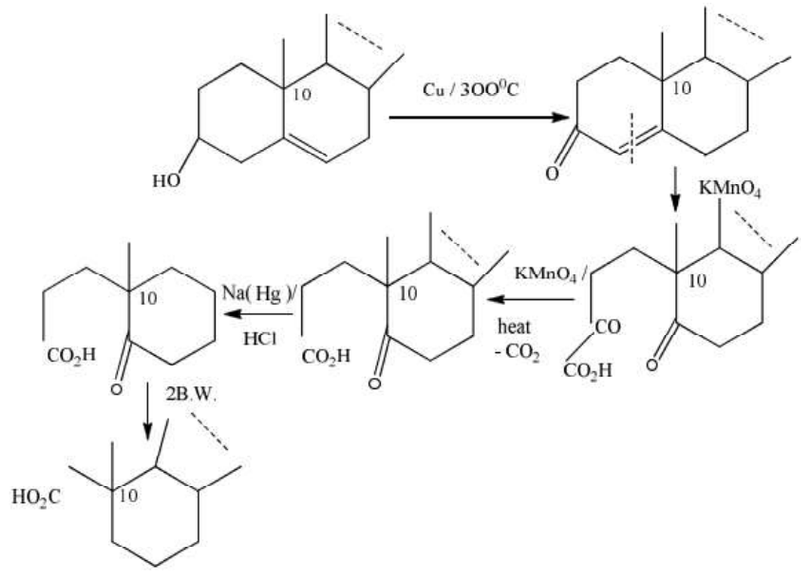


The last reactions indicates that the side chain consists of eight carbons attached with the ring D which is a five membered ring. Also, formation of Diel's hydrocarbon on heating cholesterol with selenium ,indicate that the side chain attached to the nucleus at C-17. Formation of 1,2-dimethylphenanthrene indicate that there is an angular methyl at C-13.

The presence of an angular methyl at C-10:



The carboxylic group at C-10 resists esterification and resists decarboxylation, this is due to the presence of an angular methyl group and the carbon is tertiary carbon.

Presence of an angular methyl group at C-13:

Cholesterol + H_2 / Pt $\rightarrow$ cholestanol cholestanol + CrO_3 $\rightarrow$ cholestanone cholestanone + Na (Hg) / HCl $\rightarrow$ cholestane

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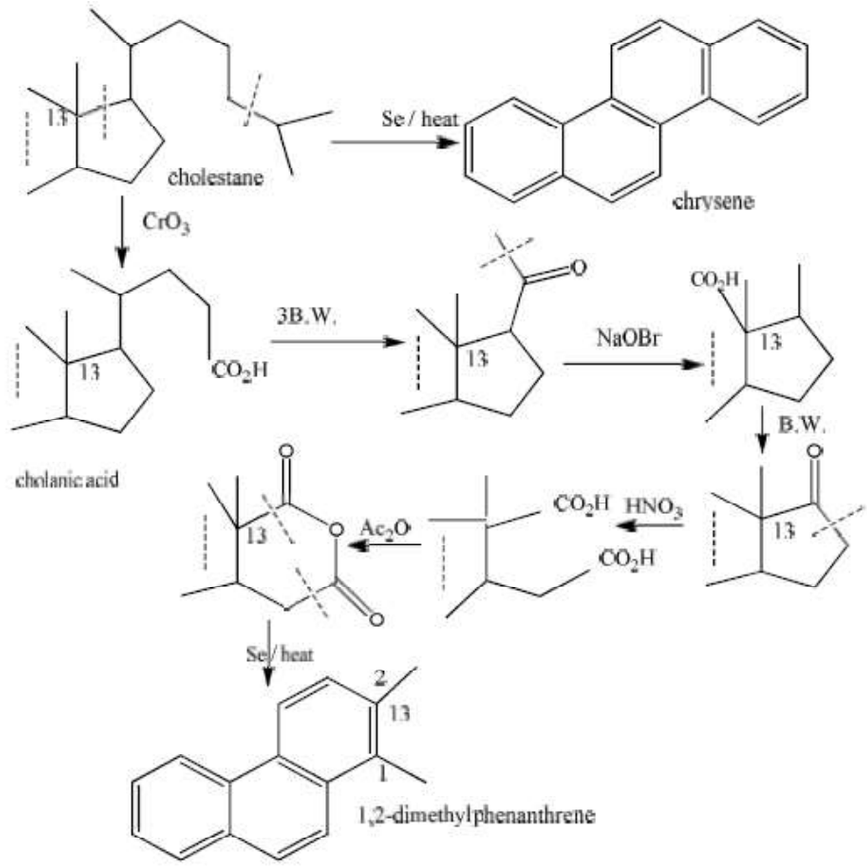

Biosynthesis of Steroids

3.3.2 Bile Acids

Bile acids are steroid acids found mostly in mammals and other vertebrates. The liver synthesizes a variety of bile acids. Bile salts are formed when bile acids are conjugated with taurine or glycine residues. Bile acids synthesized by the liver are known as primary bile acids. Bacterial activity in the colon produce secondary bile acids. The primary bile salts in humans are taurocholic acid and glycocholic acid (cholic acid derivatives) and taurochenodeoxycholic acid and glycochenodeoxycholic acid (chenodeoxycholic acid derivatives). In terms of concentration, they are roughly equal. Derivatives of cholic, chenodeoxycholic, and deoxycholic acids, as well as their 7-alpha-dehydroxylated derivatives, deoxycholic acid and lithocholic acid, are also detected, with cholic, chenodeoxycholic, and deoxycholic acids accounting for about 90% of human biliary bile acids.

Bile salts are a broad class of compounds made up of a four-ringed steroid structure, a five- or eight-carbon side chain terminating in a carboxylic acid, and multiple hydroxyl groups, the number and orientation of which varies depending on the bile salt. From the farthest to the closest to the side chain with the carboxyl group, the four rings are called A, B, C, and D. The D-ring is one carbon smaller than the others. A is usually drawn on the left, while D is drawn on the right. The

hydroxyl groups can be arranged in one of two ways: up (or out), known as beta, or down, known as alpha.

The 3-hydroxyl group in all bile acids comes from the parent molecule, cholesterol, where the 3-hydroxyl is beta. The steroid skeleton's ring lettering (left) and atom numbering (right) are recommended by IUPAC. A sterane core is formed by the four rings A-D. The enzymatic addition of a 7α hydroxyl group by cholesterol 7α-hydroxylase (CYP7A1) to generate 7α-hydroxycholesterol is the first step in the traditional pathway of hepatic bile acid production. After that, 7α-hydroxy-4-cholesten-3-one is formed. Bile acid production is broken down into several phases, requiring a total of 14 enzymes. As a result, the junction between the first two steroid rings (A and B) is altered, bending the molecule; the 3-hydroxyl is transformed to the α orientation during this process.

The simplest 24-carbon bile acid has two hydroxyl groups at the positions 3α and 7α. This is 3α, 7α-dihydroxy-5α-cholan-24-oic acid, or chenodeoxycholic acid as it is more often known. The name "cheno" comes from the fact that this bile acid was first isolated from the domestic goose. The 5α in the name refers to the orientation of the steroid nucleus's rings A and B. (in this case, they are bent). The name "cholan" refers to a steroid with 24 carbons, while "24-oic acid" refers to the carboxylic acid's location at position 24, at the end of the side-chain. Many species produce chenodeoxycholic acid, which is the prototypical functional bile acid.

3.3.3 Androsterone

Androsterone is an endogenous steroid metabolite, neurosteroid, and putative pheromone derived from testosterone and DihydroTestosterone (DHT), which displays weak androgenic properties. In testes, it is formed from progesterone. Androsterone sulfate is clinically recognized as one of the major androgen metabolites found in urine, in male and females.

Functions of Androsterone

It is responsible for male characteristics such as:

- Male sexual and reproductive function.
- Development of secondary sexual characteristics in men, including facial and body hair growth voice change and texture of skin.
- It also affects bone and muscle development, and metabolism.

Isolation

It was first isolated by Adolf Friedrich Johann Butenandt et al in 1931 from male urine (about 15 mg from 15,000 litres of urine). In urine, it is found as sulphate. As a first step, urine is concentrated and acid is added into it and then it is extracted with chloroform by adding potassium hydroxide. All the acidic and phenolic impurities are remove. After removing the impurities, Grignard reagent is added and it forms the Grignard derivative. Hormones are isolated by chromatographic technique.

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Constitution

The constitution of androsterone is:

- The molecular formula of androsterone is $C_{19}H_{30}O_2$.
- The melting point of androsterone is 184-185°C and optical rotation is $[\alpha]_D$ + 94°.
- Presence of tetracyclic: The general formula for androsterone is C_nH_{2n}–6.

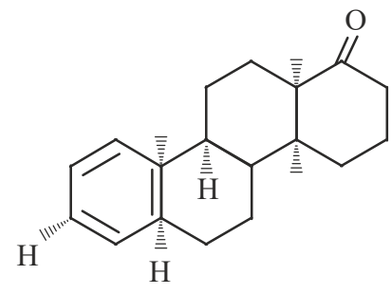
  The calculating double bond is

  $$\text{DBE of } C_{19}H_{30}O_2 = 19 + 1 - \frac{30}{2} = 5$$

  (1 double bond due to C = O and so there are four rings.)
- Androsterone behaves as a saturated compound, and since it forms mono-esters by acetylation.
- With benzoylation, androsterone forms mono-benzoyl derivative. This shows the presence of one hydroxyl group.
- The functional nature of the other oxygen atom was shown to be Oxo. Since androsterone forms a monoxime with hydroxyl amine, it indicates the presence of one ketonic group.

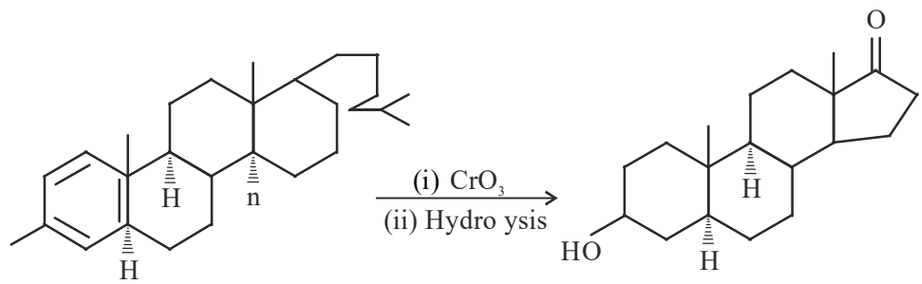
$$C_{17}H_{28}\begin{bmatrix}CHOH\\C=O\end{bmatrix}\xrightarrow{NH_2OH}C_{17}H_{28}\underbrace{\begin{bmatrix}CHOH\\C=N-OH\end{bmatrix}}_{\text{Monoxime}}$$

- Butenandt (1932) therefore proposed a structure for androsterone:

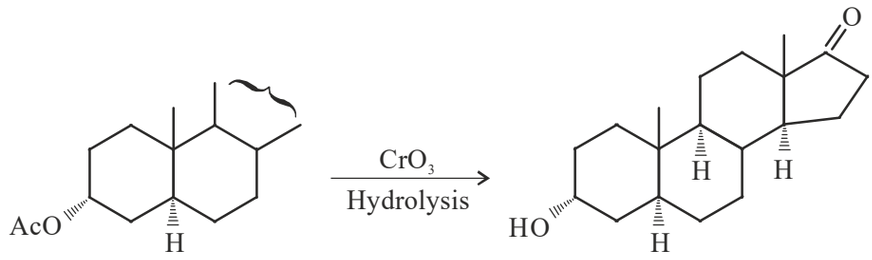


Synthesis of Androsterone

- Ruzicka (1934) proposed the following synthesis:



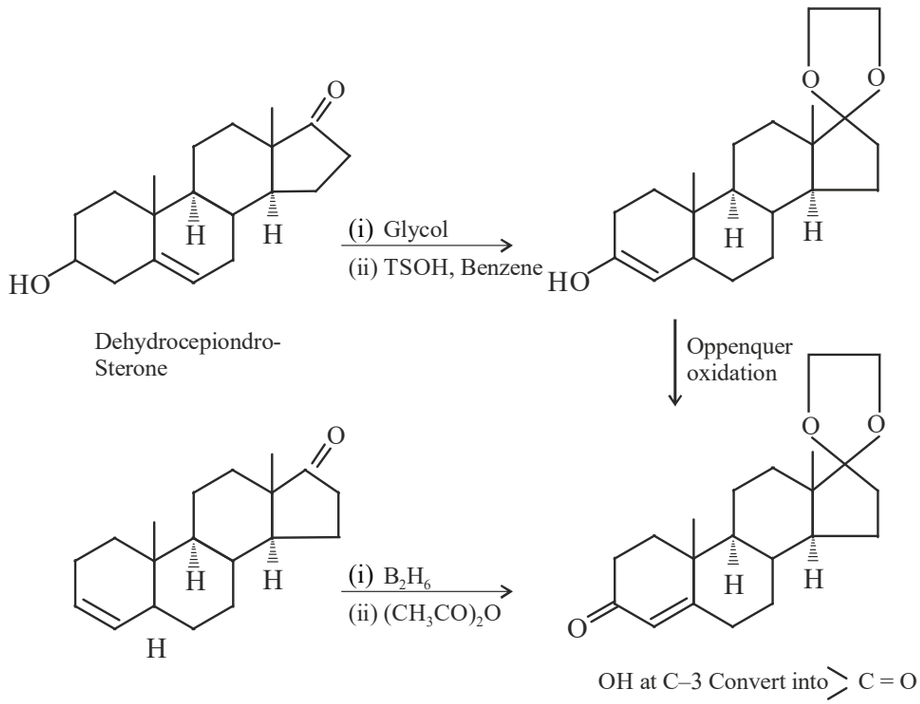
5α-Chlalestanyl 3β-acetate — Epiandrosterone



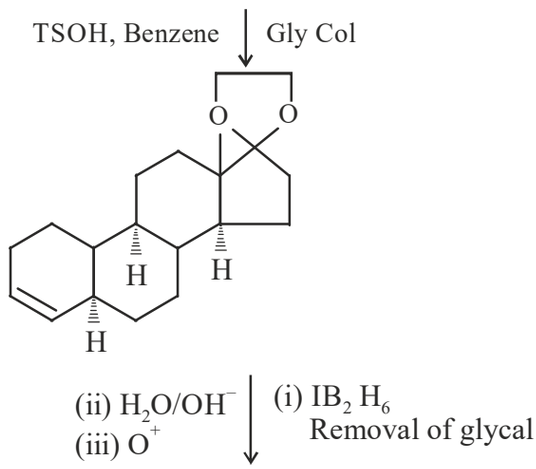
5α-Chlalestanyl 3α-acetate

Androsterone

- Caglito synthesis (1964)



Dehydrocepiondro-Sterone



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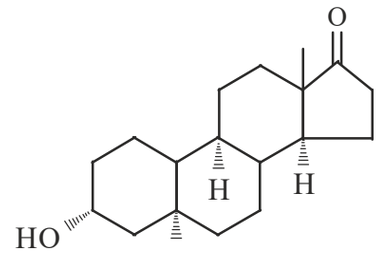


Androsterone

3.3.4 Testosterone ($C_{19}H_{28}O_2$)

Testosterone is a primary male sex hormone. It is essential in the development of man reproductive tissue as well as increase muscle, bone and growth of body hair. It is the natural androgen secreted by the interstitial cell. Testosterone was isolated by the team of biochemist Ernst Laqueur (1935) from testes.

Constitution

The constitution of testosterone is:

- Molecular formula of testosterone is $C_{19}H_{28}O_2$ on the basis of analytical data.
- On catalytic reduction testosterone takes two molecules of hydrogen, because one is used for conversion ketone group into secondary alcoholic group and second hydrogen molecule must be used to convert double bond to saturate it.

$$C_{17}H_{26}\begin{bmatrix}CHOH\\C=O\end{bmatrix}\xrightarrow{2H_2}C_{17}H_{28}\underset{\text{Saturated diol}}{\begin{bmatrix}CHOH\\CHOH\end{bmatrix}}$$

- Presence of tetracyclic structure: The molecular formula of parent hydrocarbon of testosterone corresponds to the general formula C_nH_{2n-6}. Therefore, testosterone should be tetracyclic.
- Testosterone behaves as a saturated compound, and since it forms monoester with acetic anhydride by acetylation. Formation of monoester indicates the presence of alcoholic group.

$$C_{17}H_{28}O_2\xrightarrow{(CH_3CO)_2O}C_{17}H_{26}\begin{vmatrix}>CHOCOCH_3\\>C=O\end{vmatrix}$$

- On oxidation it gives diketone

$$C_{17}H_{28}O_2\xrightarrow{[O]}\underset{\text{diketone}}{C_{17}H_{26}\begin{vmatrix}>C=O\\>C=O\end{vmatrix}}$$

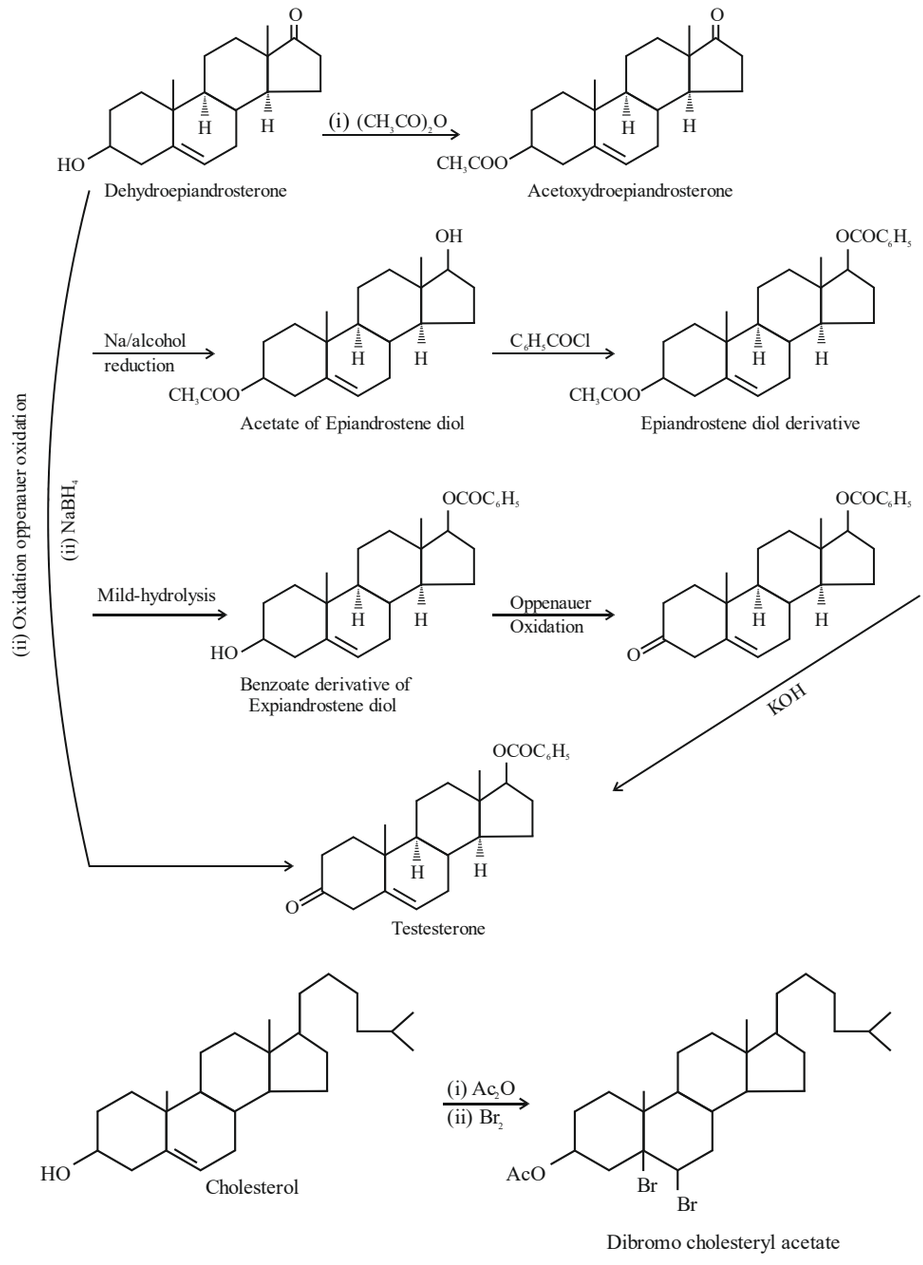
- Testosterone forms monoxime with hydroxyl amine, which indicates the presence of ketonic group in the molecule.

$$C_{17}H_{28}O_2 \xrightarrow{NH_2OH} C_{17}H_{26}\begin{vmatrix}CHOH\\ >C=N-OH\end{vmatrix}$$
oxime

Synthesis of Testosterone

Following are the methods of synthesis of testosterone:

- Ruzicka and Butenandt Synthesis (1935):



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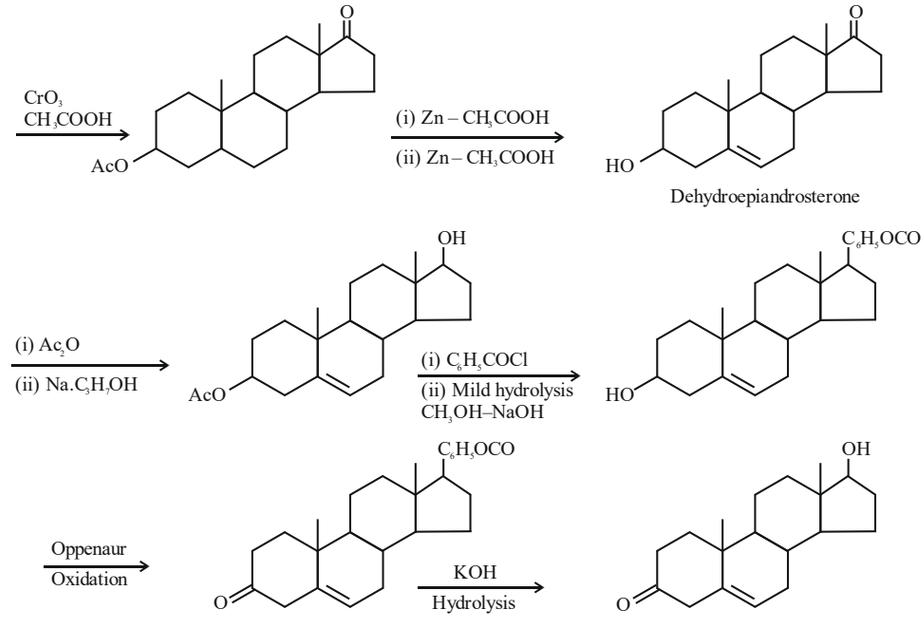
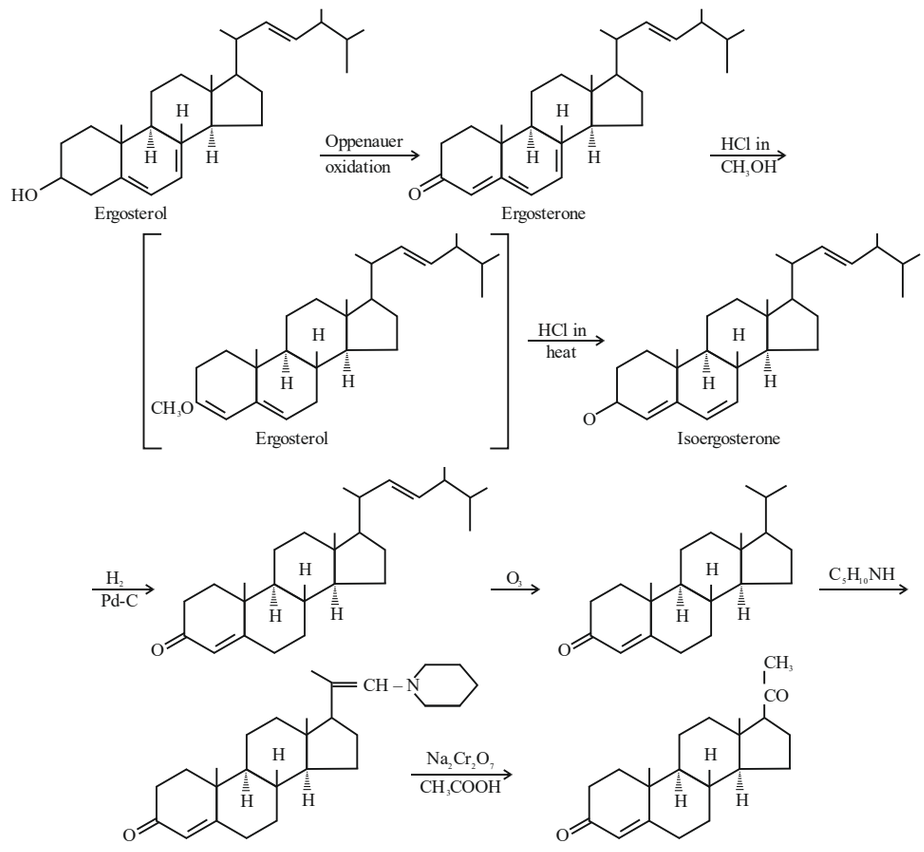
- Mamali Synthesis (1938):



Synthesis method



3.3.5 Progesterone $C_{21}H_{30}O_2$

Progesterone hormones are chemical messengers produced in the ovaries, placenta and adrenal glands; it is a steroid hormone that prepares the uterus for a fertilized ovum and maintains pregnancy. It also plays an important role in brain function as a neurosteroid. It was first isolated in pure form by Willard M. Allen and George W. Corner from the corpora lutea of pregnant cows.

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Properties

Progesterone is a stable white crystalline solid with two polymorphic forms, i.e., prism form and needle form. The prism form (M.P. 128°C) is obtained on slow crystallisation from aqueous alcohol and the needle form (M.P. 121°C) is obtained from petroleum ether.

Constitution of Progesterone

The constitution of progesterone is discussed below:

- **Molecular Formula:** The molecular formula of progesterone has been found to be $C_{21}H_{30}O_2$.
- **Presence of Ketonic Group:** Progesterone on treatment with hydroxylamine forms dioxime derivative, thus shows that it contains two ketonic groups.
- **Presence of Double Bond:** When catalytically reduced, it takes up three molecules of hydrogen to form the dialcohal $C_{21}H_{36}O_2$. As two molecules of hydrogen are utilized for the conversion of two ketonic groups, into two secondary alcoholic groups, the third molecule of hydrogen is added on the double bond to saturate it. Thus, progesterone must contain one double bond.
- **Presence of Steroid Nucleus:** Progesterone contains two ketonic groups and one double bond, it follows that the parent hydrocarbon of progesterone is $C_{21}H_{36}$. But this general formula corresponds to the general formula C_nH2_{n-6}. There, it means that progesterone is tetracyclic:

$$\text{DBE} = \frac{C+1-H}{2} \text{ i.e. } \frac{21+1-36}{2} = 4 \text{ rings.}$$

- **Presence of α, β-Unsaturated Ketonic Group:** Progesterone is sensitive to alkalies, it means that it contains an α, β-unsaturated ketonic group. Further the absorption spectrum (λ_{max} 240nm) of progesterone confirms this grouping

- **Presence of $CH_3-\overset{O}{\overset{\|}{C}}-$ Group:**

  When progesterone is heated with one halogen and NaOH it yeilds haloform, i.e., it undergoes haloform reaction. This reaction indicate that progesterone contains group.

  On the basis of the above facts, the following structure formula has been assigned to progesterone.

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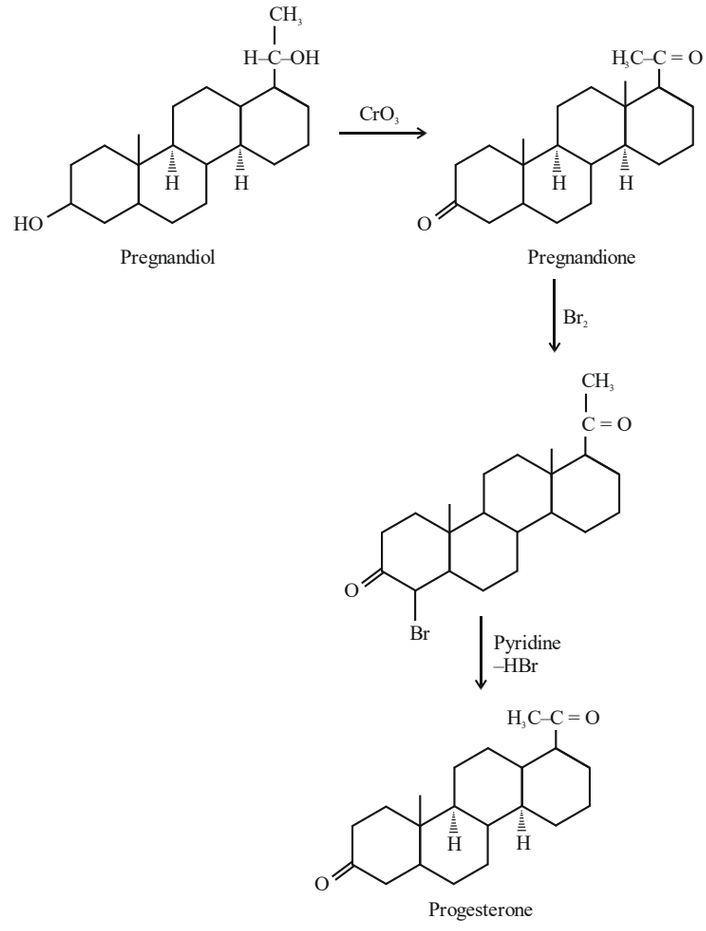
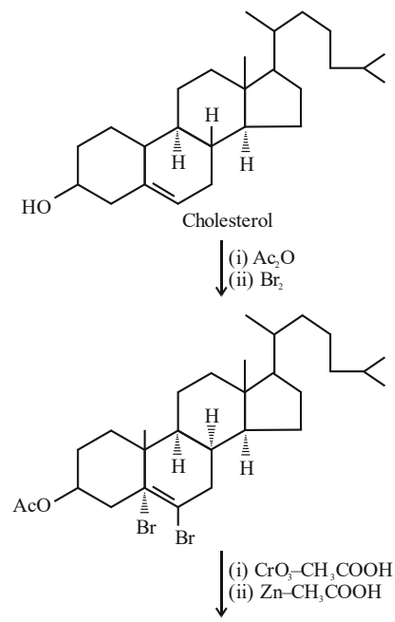
Synthesis of progesterone

- By pregnandiol (Butenandt et.al 1934)



- By Progesterone form Cholesterol



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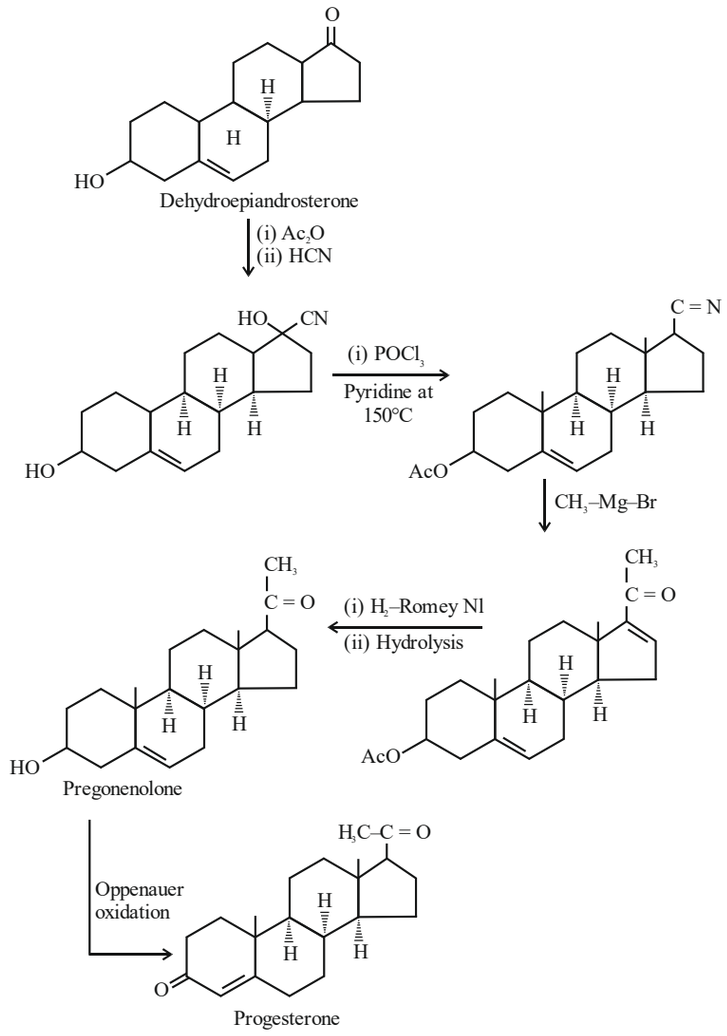
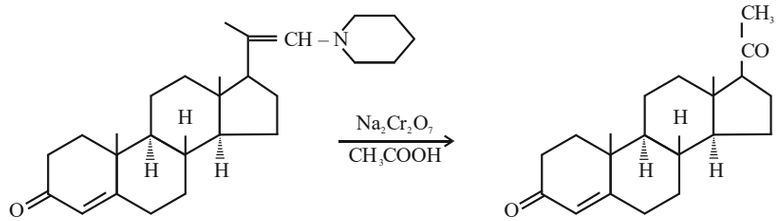


- Progesterone from ergosterol (Shephord Synthesis 1955):



3.3.6 Estrone

Estrone (E1), also spelled oestrone, is a steroid, a weak estrogen, and a minor female sex hormone. It is one of three major endogenous estrogens, the others being estradiol and estriol. In order to isolate estrone from dates seeds, the seeds are grounded in a pulverizing mill, and is refluxed with 3 N hydrochloric acid for 3 hours. After filtration and washing with water, the dried filter cake is extracted in a Soxhlet apparatus with 3 litres of methanol for 24 hours, then 3 litres of acetone for 24 hours, and finally with 3 litres of water for 24 hours. All of the solvents are

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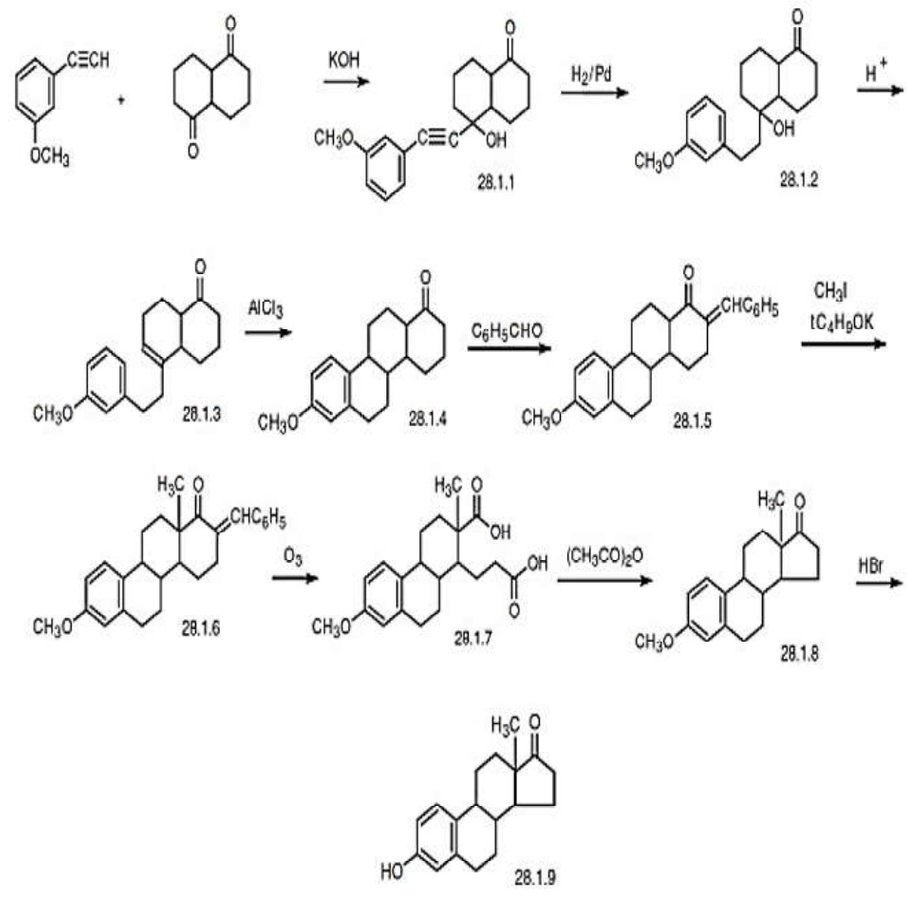
collected separately. After vacuum distillation, estrone is collected and identified with the help of several spectral techniques.

Biosynthesis of estrone is done from cssue. The IUPAC name of estrone is 3-hydroxy estra-1,3,5 (10)-trien-17-one. It is made synthetically in various ways. This synthesis is carried out in the following manner.

In the first step, condensation of 3-methoxy phenyl acetylene with bicyclohexane-1,5-dione in a Favorskii reaction forms carbinol. Triple bond is reduced by hydrogen in the presence of palladium form tertiary alcohol, which was then dehydrated in acidic medium to give the compound. Intramolecular alkylation of this compound in the presence of anhydrous aluminium chloride formed a tetra cyclic ketone which during condensation with benzaldehyde was transformed into an eneone.

This was methylated at the β-position relative to the keto-group by methyl iodide in the presence of potassium tert-butylate, and the resulting compound under ozonolysis, forming the dicarboxylic acid. Cyclization of this compound to a cyclopentanone derivative lead to the formation of methyl ester of the desired estrone and demethylation of the phenolic hydroxyl group by hydrobromic acid formed the desired estrone.



3.3.7 Aldosterone

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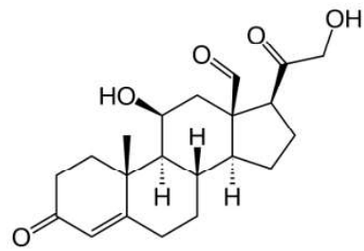
The Aldosterone is a steroid hormone that is secreted by the adrenal glands; it is characterized by the presence of an aldehyde functional group in carbon 18. It was isolated for the first time in 1953 and later synthesized in the laboratory by Derek Barton. Also, this hormone is in the group of mineralocorticoids, which are produced in the adrenal cortex which is also responsible for the manufacture of glucocorticoids. In addition, aldosterone is secreted into the glomerular zone, which is the outermost, thin layer of the cortex.

The main function of aldosterone is to regulate mineral metabolism by facilitating the reabsorption of sodium in the kidneys, although it is also responsible for eliminating potassium.



Isolation of aldosterone first occurred in 1953, which means that its presence was not unknown before it was given a common name within the official nomenclature. However, it was not until later that the British scientist Derek Harold Richard Barton found a way to synthesize this hormone in controlled environments, that is, in the premises of his laboratory.

At this point, it is not surprising that Barton understood the nature of aldosterone well enough to win the Nobel Prize for Chemistry in 1969 with Norwegian physical chemist, Odd Hassel.

Synthesis

Aldosterone is synthesized in the body from corticosterone, which is a steroid derived from cholesterol. Production of aldosterone (in adult humans, about 20–200 micrograms per day) in the zona glomerulosa of the adrenal cortex is regulated by the renin-angiotensin system. Renin is secreted from the kidneys in response to variations in blood pressure, volume, plasma sodium and potassium levels. Renin acts on a protein circulating in the plasma called angiotensinogen, cleaving this substance into angiotensin I. Angiotensin I is subsequently converted to angiotensin II, which stimulates the release of aldosterone from the adrenal glands.

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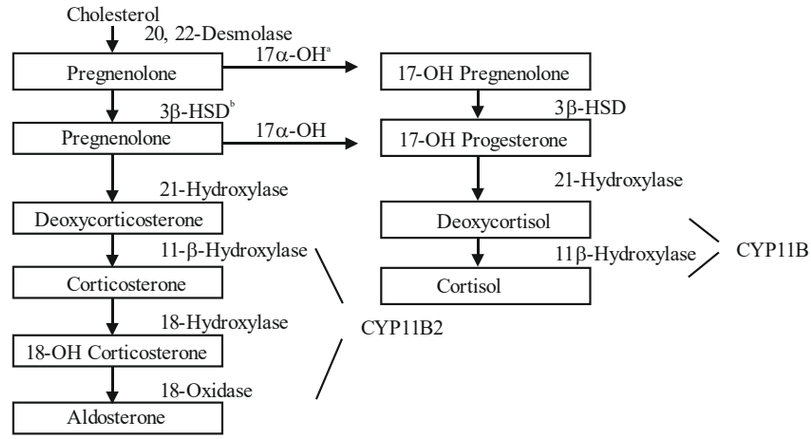

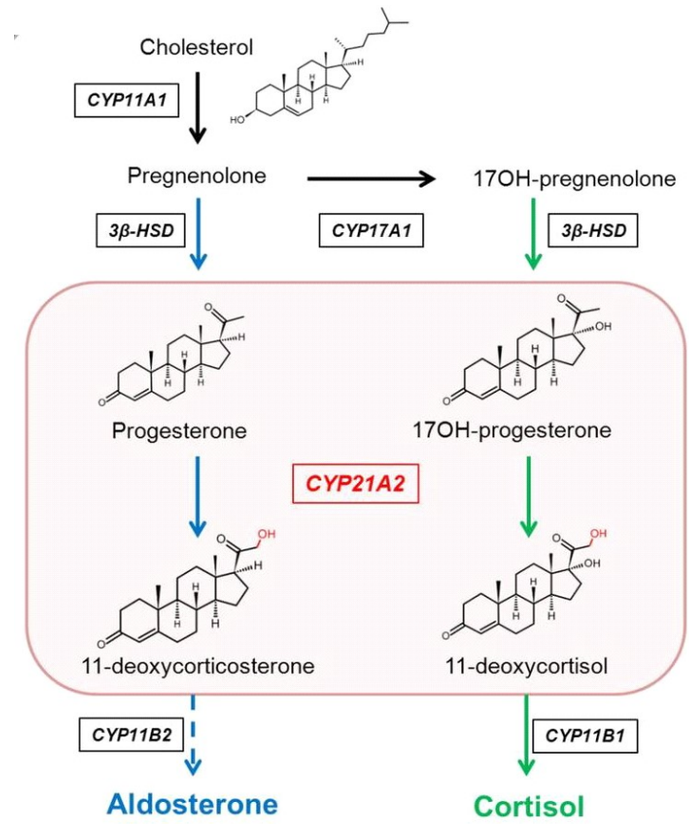

Check Your Progress

3. What are the primary bile salts in humans?
4. What are the functions of androsterone?
5. Who isolated aldosterone for the first time?

3.4 ANSWERS TO CHECK YOUR PROGRESS QUESTIONS

NOTES

1. The steroids have two principal biological functions: as important components of cell membranes which alter membrane fluidity; and as signalling molecules.
2. Stereochemistry of the steroids can be defined as the spatial arrangement of the substituents of steroids.
3. The primary bile salts in humans are taurocholic acid and glycocholic acid (cholic acid derivatives) and taurochenodeoxycholic acid and glycochenodeoxycholic acid (chenodeoxycholic acid derivatives).
4. The androsterone is responsible for male characteristics such as:
   a. Male sexual and reproductive function.
   b. Development of secondary sexual characteristics in men, including facial and body hair growth voice change and texture of skin.
   c. It also affects bone and muscle development, and metabolism.
5. Aldosterone was isolated for the first time in 1953 and later synthesized in the laboratory by Derek Barton.

3.5 SUMMARY

- Steroid refers to any of a class of natural or synthetic organic compounds characterized by a molecular structure of 17 carbon atoms arranged in four rings.
- The steroid group includes all the sex hormones, adrenal cortical hormones, bile acids, and sterols of vertebrates, as well as the molting hormones of insects and many other physiologically active substances of animals and plants.
- Steroids vary from one another in the nature of attached groups, the position of the groups, and the configuration of the steroid nucleus (or gonane).
- Natural steroids are compounds that mimic the steroids that human bodies naturally produce, such as, the hormones testosterone, progesterone, etc.
- Steroids vary by the functional groups attached to this four-ring core and by the oxidation state of the rings. Sterols are forms of steroids with a hydroxy group at position three and a skeleton derived from cholestane.
- The steroids are a family of compounds widely distributed in plants and animals. Common to the structure of all compounds of this class is a tetracyclic framework composed of the phenanthrene nucleus to which is fused at the 1, 2-positions a cyclopentene ring.
- Gonane, also known as steran or cyclopentanoperhydrophenanthrene, the simplest steroid and the nucleus of all steroids and sterols, is composed of seventeen carbon atoms in carbon-carbon bonds forming four fused rings in a three-dimensional shape.

NOTES

- Cholesterol refers to a group of organic molecules. A cholesterol is a form of lipid known as a sterol (or modified steroid). All animal cells produce cholesterol, which is an important structural component of cell membranes.
- Bile acids are steroid acids found mostly in mammals and other vertebrates. The liver synthesizes a variety of bile acids. Bile salts are formed when bile acids are conjugated with taurine or glycine residues.
- Androsterone is an endogenous steroid metabolite, neurosteroid, and putative pheromone derived from testosterone and dihydrotestosterone (DHT), which displays weak androgenic properties.
- Testosterone is a primary male sex hormone. It is essential in the development of man reproductive tissue as well as increase muscle, bone and growth of body hair. It is the natural androgen secreted by the interstitial cell.
- Progesterone hormones are chemical messengers produced in the ovaries, placenta and adrenal glands; it is a steroid hormone that prepares the uterus for a fertilized ovum and maintains pregnancy.
- The Aldosterone is a steroid hormone that is secreted by the adrenal glands; it is characterized by the presence of an aldehyde functional group in carbon 18.

3.6 KEY TERMS

- **Steroid:** A steroid is a biologically active organic compound with four rings arranged in a specific molecular configuration.
- **Cholesterol:** Cholesterol refers to a group of organic molecules. A cholesterol is a form of lipid known as a sterol (or modified steroid).
- **Gonane**: Gonane are the simplest steroid and the nucleus of all steroids and sterols, and is composed of seventeen carbon atoms in carbon-carbon bonds forming four fused rings in a three-dimensional shape.
- **Adrenal Cortex Hormones:** The outer portion of the adrenal gland located on top of each kidney is called adrenal cortex, which produces steroid hormones which regulate carbohydrate and fat metabolism and mineralocorticoid hormones which regulate salt and water balance in the body.
- **Nomenclature:** It is a system of names or terms, or the rules for forming these terms in a particular field of arts or sciences.

3.7 SELF ASSESSMENT QUESTIONS AND EXERCISES

Short Answer Questions

1. Write a short note on the stereochemistry of steroids.
2. What is the constitution of androsterone?
3. What do you mean by esterone? How is it synthesized?

Long Answer Questions

1. Discuss the occurrence, nomenclature and basic skeleton of steroids.
2. Comment on the constitution and synthesis of progesterone.
3. Discuss how aldosterone is isolated and synthesized.

NOTES

3.8 FURTHER READING

Rasheed, Anas and M. Qasim. 2013. *A Review of Natural Steroids and Their Applications*. Germany: Lap Lambert Academic Publishing GmbH KG.

Lednicer, D. 2011. *Steroid Chemistry at a Glance*. Germany: Wiley.

Torgov, I. V. 1965. *Achievements in the Total Synthesis of Natural Steroids.* (n.p.): Verlag nicht ermittelbar.

Weisbart, M. 1973. *Isolation and Purification of Hormones*. United States: MSS Information Corporation.

UNIT 4 PLANT PIGMENTS

NOTES

Structure

4.0 INTRODUCTION

Plant pigments or phytochromes are the substances produced by plants that have a colour resulting from selective colour absorption. The primary function of pigments in plants is photosynthesis, which uses the green pigment chlorophyll and several colorful pigments that absorb as much light energy as possible.

There are many different plant pigments, and they belong to different classes of organic compounds. Plant pigments give colour to leaves, flowers, and fruits and are also important in controlling photosynthesis, growth, and development.

Pigments play an important role in the pollination as well where pigment accumulation or loss can lead to change in the floral colour, signalling to pollinators which flowers are rewarding and contain more pollen and nectar. Plant pigments include many molecules, such as, porphyrins, carotenoids, anthocyanins and betalains. All biological pigments selectively absorb certain wavelengths of light while reflecting others.

In this unit, you will study about nomenclature, structure, function and synthesis of various plant pigments and biosynthesis of flavonoids.

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4.1 OBJECTIVES

After going through this unit, you will be able to:

- Understand the occurrence of the various plant pigments
- Conceptualize the nomenclature and structure plant pigments
- Classify the plant pigments
- Explain the structure of plant pigments
- Demonstrate the synthesis of plant pigments
- Elaborate on the Biosynthesis of Flavonoids

4.2 OCCURRENCE, NOMENCLATURE, STRUCTURAL DETERMINATION, ISOLATION AND SYNTHESIS OF VARIOUS PLANT PIGMENTS

There are different type of pigments that a plant can have. The variation in plant pigments are due to the different wavelength of light they absorb. Plant pigments are used to manufacture natural dyes. Chlorophyll is the most abundant plant pigment in the world. Inplants carotenoids, flavonoids and betalains are also a green pigment. Some plants having yellow pigments in them are called lutein, whereas, the red pigment in plant is lycopene anthocyanin are water soluble pigments that are present in the flower petal of different species.

Plant pigments are important in controlling photosynthesis, growth and development of plants. Pigments also protect plant from damage caused by UV (Ultra Violet) and visible light.

4.2.1 Apigenin ($C_{15}H_{10}O_5$)

IUPAC Name: 5, 7, 4′–Trihydroxy Flavone

Structure

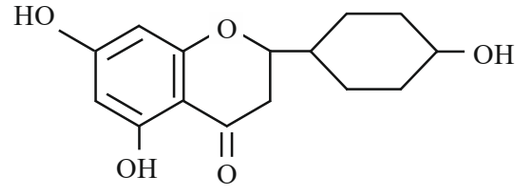

Fig 4.1 *Apigenin*

Constitution

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1. **Molecular Formula:** On the basis of analytical data, the molecular formula is $C_{15}H_{10}O_5$.
2. Alcoholic solution of apigenin gives pink colour with Mg and dilute HCl.
3. It shows absorption maxima λ_{max} at 267nm (bond II) and 336V nm (bond I) in UV-spectrum.
4. On acetylation with acidic anhydride and sodium acetate gives tri-acetyl derivative, indicating the presence of three hydroxyl groups.

$$C_{15}H_7O_2(OH)_3 \xrightarrow[CH_3COONa]{(CH_3CO)_2} C_{15}H_7O_2(OAC)_3$$

5. By Zeisel's method shows the absence of methoxy group. Now we see that possible structure of apigenin is

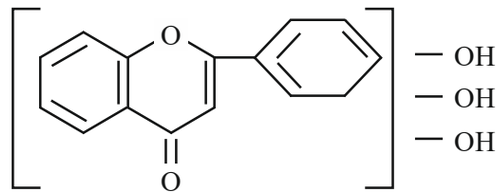
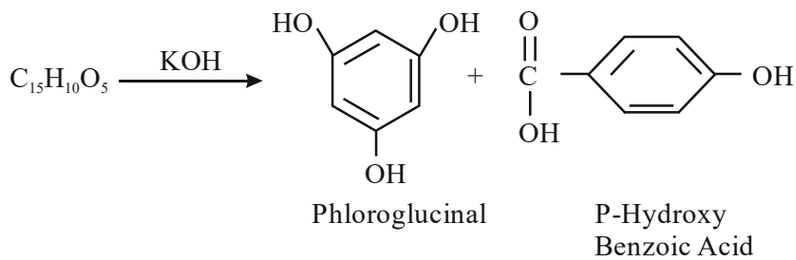


Fig. 4.2 *Structure of Apigenin*

6. **Position of Hydroxyl Group**
   (i) Tri-methoxy apigenin on oxidation with $KMnO_4$ gives p-methoxybenzoic acid as one of the products. Formation of this compound shows that ring-B contains a hydroxy group at position-IV.
   (ii) Apigenin on KOH degradation gives phloroglucinol and p-Hydroxy benzoic acid.



Formation of phloroglucinol shows that ring A contains hydroxyl group at position V and position VII similarly the formation of p-hydroxy benzoic acid shows that ring B contains a hydroxyl group at position IV.

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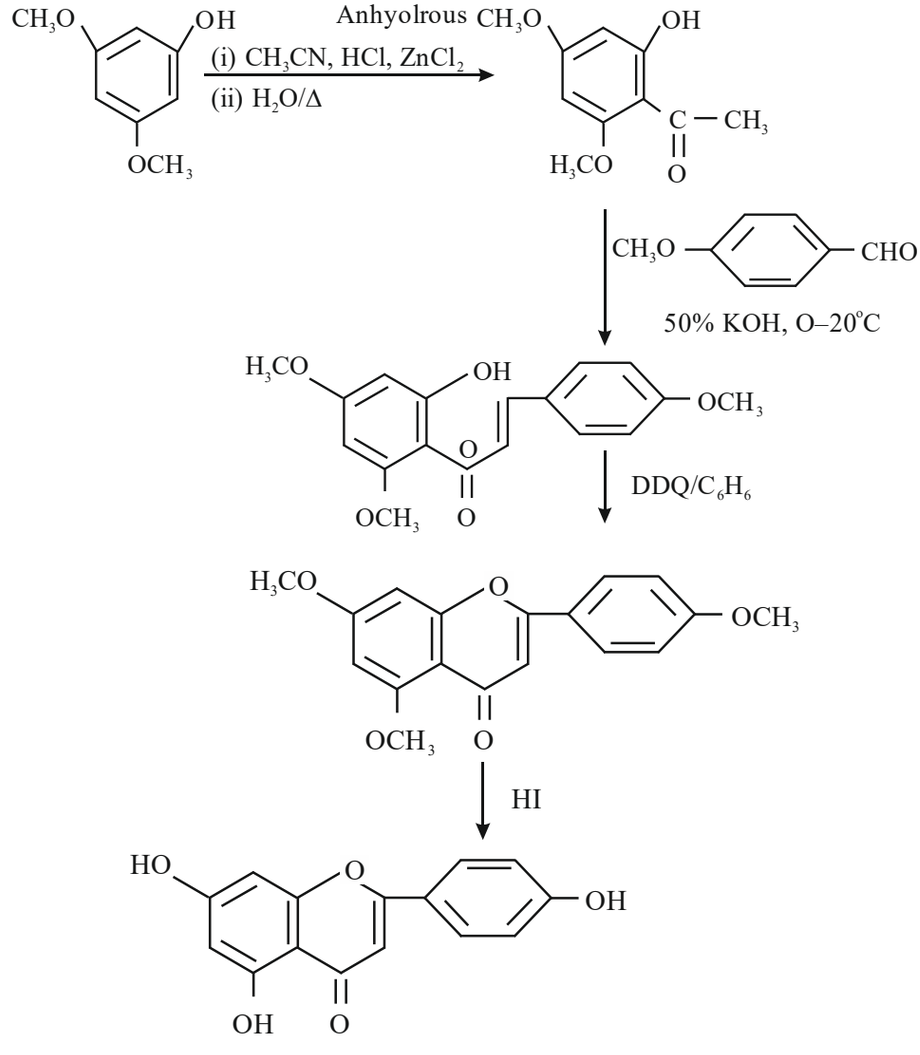
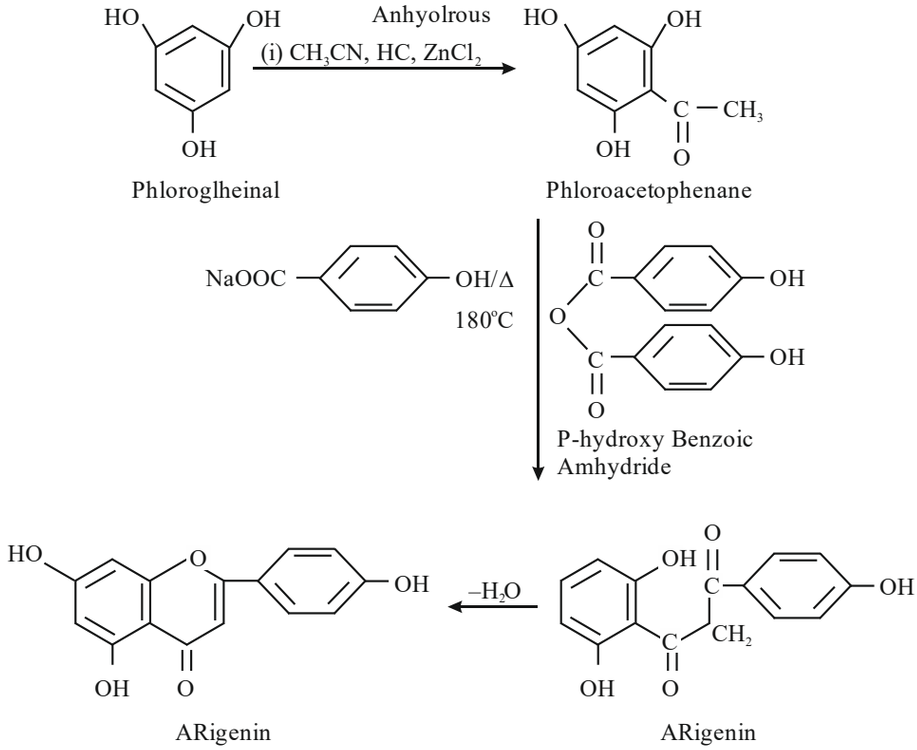
7. **Synthesis of Apigenin:** The structure of apigenin has been confirmed by its synthesis.

(i) **Chalcone Method**



Apigenin

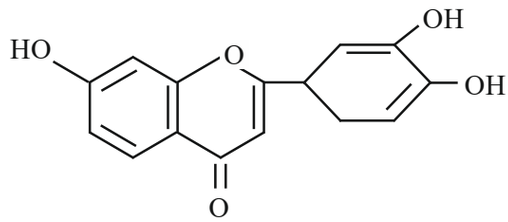
(ii) Allan-Robinson Synthesis



4.2.2 Luteolin ($C_{15}H_{10}O_6$)

IUPAC Name: 5-7-dihydroxy-2-(3, 4-dihydroxy phenyl)-chromene-4-one

Structure:



Luteolin

Fig. 4.3 *Structure of Luteolin*

Constitution

1. Molecular formula of luteolin is $C_{15}H_{10}O_6$
2. Alcoholic solution of luteolin gives dark pink colour with Mg turnings and dilute HCl.

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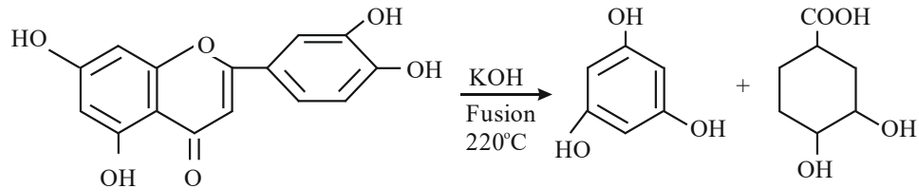
3. Luteolin on acetylation with acetic anhydride and fused sodium acetate yields tetra-acetyl luteolin, indicating the presence of four hydroxyl groups.

$$C_{15}H_6O_2(OH)_4 \xrightarrow{\text{acetylation}} C_{15}H_6O_2(OAC)_4$$

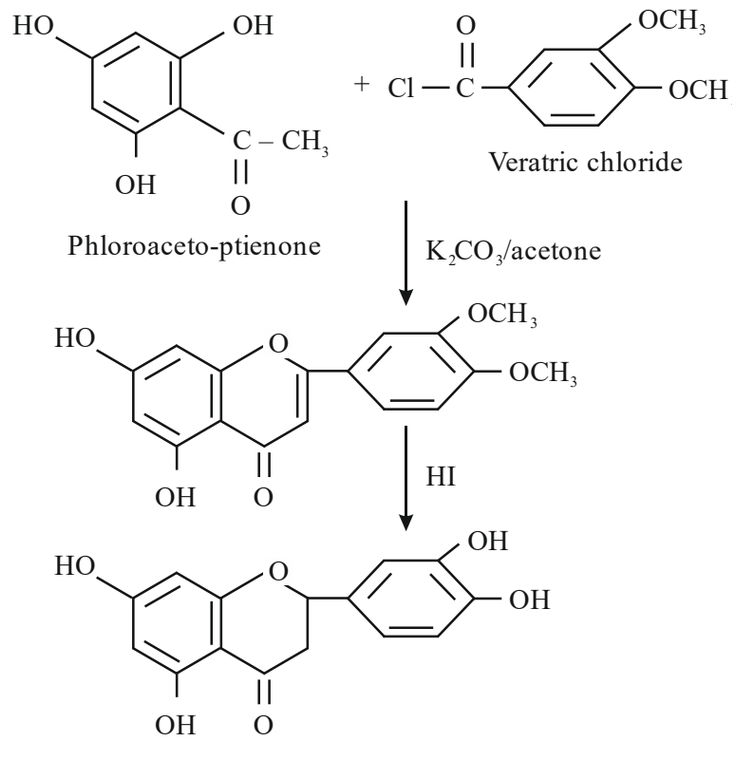
4. By Zeisels method it shows the absence of any methoxy group.
5. **Position of Hydroxyl Group:**
   (i) **Fusion with KOH:** By the process of KOH degradation, luteolin gives phloroglucinol and dihydroxy benzoic acid.



Phloroglucinal dihydroxy benzoic acid

Phloroglucinal shows that ring A contain hydroxy group at position V and VII and dihydroxyl benzoic acid shows the presence of Hydroxyl groups at position III and IV in ring B.

6. Synthesis of Luteoline (Baker-Venkataraman synthesis)



4.2.3 Quercetin ($C_{15}H_{10}O_7$)

IUPAC Name: 2-(3, 4-dihydroxy phenyl) -3, 5, 7 trihydroxy chromen-4-one

Structure:

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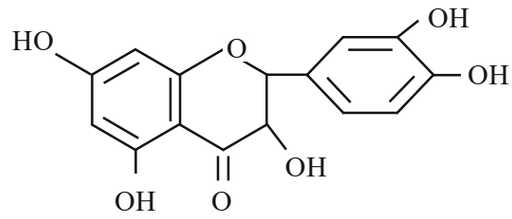


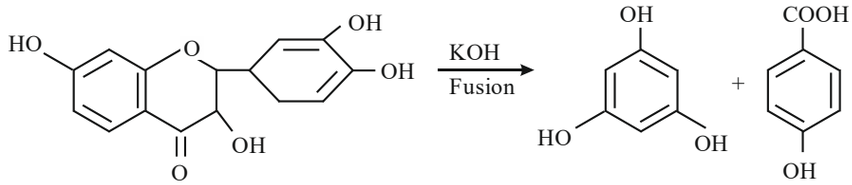
Fig. 4.4 *Structure of Quercetin*

Constitution

1. Molecular formula of quercetin is $C_{15}H_{10}O_7$.
2. Quercetin on hydrolysis gives quercetin and rhamnose sugar

$$C_{21}H_{20}O_{11} + H_2O \xrightarrow[HCl]{acid} \underset{\text{quercetin}}{C_{15}H_{10}O_7} + \underset{\text{rhamnose sugar}}{C_6H_{12}O_6}$$

3. Position of Hydroxyl group
   (i) On acetylation, quercetin forms penta-acetyl derivative.
   (ii) On methylation, quercetin forms penta-methyl derivatives.
   (iii) The phenolic nature of hydroxyl group is determined by the solubility of quercetin in alkali solution.
   (iv) **Fusion with KOH:** By the process of KOH degradation, quercetin gives phloroglucinol and protocatechuic acid.



Quecocetin Phlroglucinol Protocatechuic acid

4. **Synthesis of Quercetin**
   (i) **Robinson synthesis (1926):** w-methoxy phloroacetophenone is condensed with veratric anhydride in the presence of potassium salt of veratric acid.

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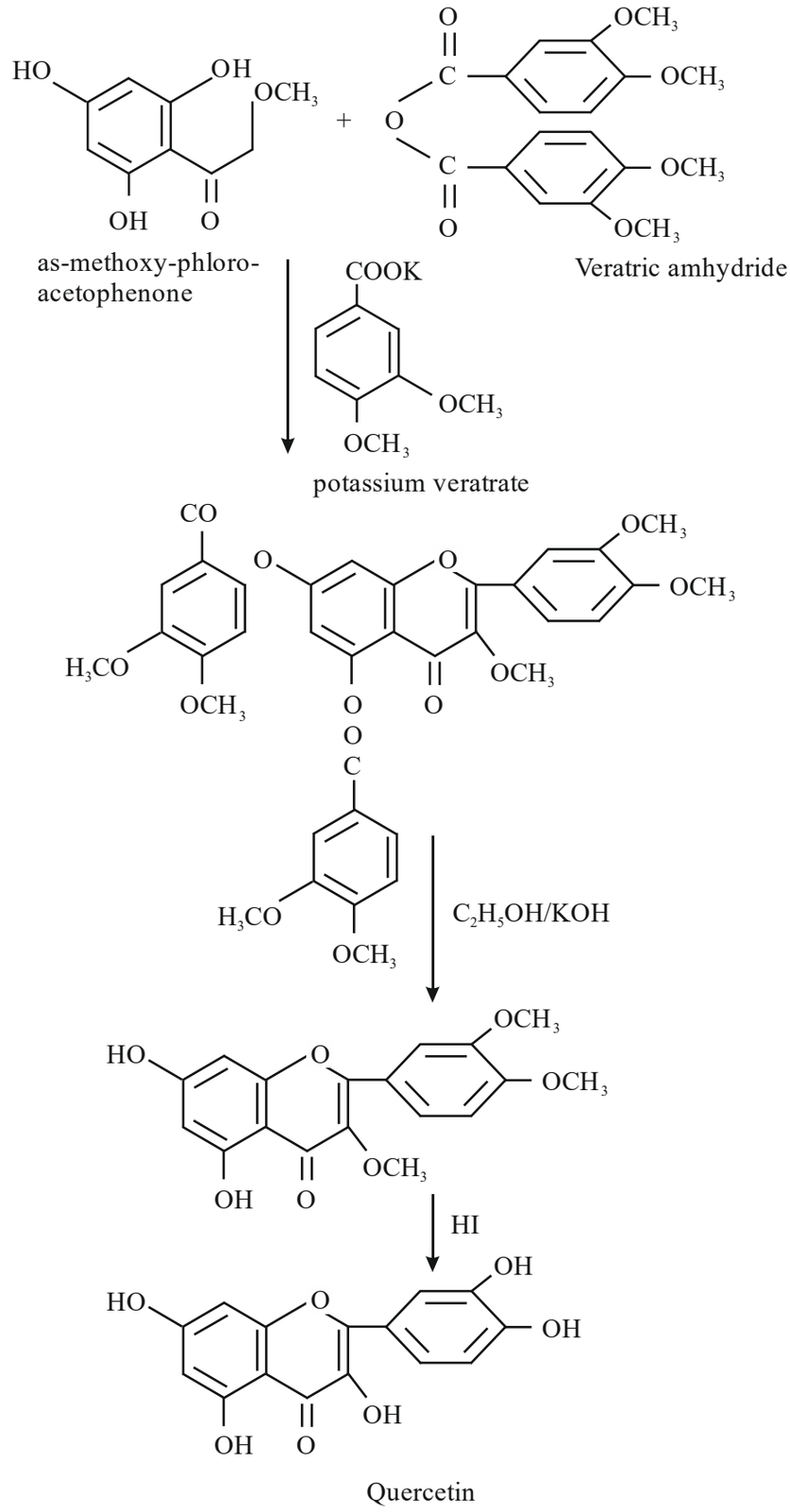
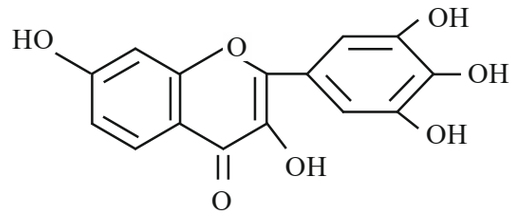

HO
OH
OCH3
OH
O
+
O
OCH3
C
OCH3
O
C
OCH3
O
OCH3
as-methoxy-phloro-acetophenone
Veratric amhydride
COOK
OCH3
OCH3
potassium veratrate
CO
H3CO
OCH3
O
O
OCH3
OCH3
O
O
O
C
H3CO
OCH3
C2H5OH/KOH
HO
O
OCH3
OCH3
OH
O
OCH3
HI
HO
O
OH
OH
OH
OH
O
Quercetin

4.2.4 Myricetin ($C_{15}H_{10}O_8$)

IUPAC Name: 3, 5, 7-trihydroxy-2- (3, 4, 5-trihydroxyphenyl)-4 H-1-benzopyram-4-one

Structure



Myricetin

Fig. 4.5 *Structure of Myricetin*

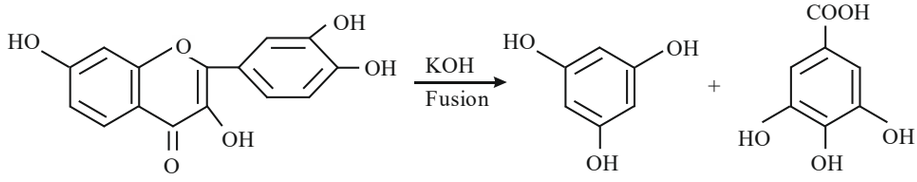
Constitution

1. Molecular formula of myricetin is $C_{15}H_{10}O_8$.
2. It shows absorption maxima λ_{max} at 350-390 nm (band II) and 250-270 nm (band I) in UV-spectrum.
3. Alcoholic solution of myricetin gives magenta colour with magnesium and dilute HCl.
4. **Position of Hydroxyl Group**
   (i) On acetylation, myricetin gives hexa-acetate derivative. Myricetin on methylation gives hexamethyl myricetin

$$\underset{\text{Myricetin}}{C_{15}H_4O_2(OH)_6} \xrightarrow[CH_3COCl]{AC_2O} \underset{\text{Hexa acetate derivative}}{C_{15}H_4O_2(OAC)_6}$$

$$\underset{\text{Myricetin}}{C_{15}H_4O_2(OH)_6} \xrightarrow{(CH_3)_2SO_4} \underset{\text{Hexa amethyl myricetin}}{C_{15}H_4O_2(OCH_3)_6}$$

   (ii) **Fusion with KOH:** Myricetin on KOH degradation gives phloroglucinol and gallic acid.



phloroglucinal gauic acid

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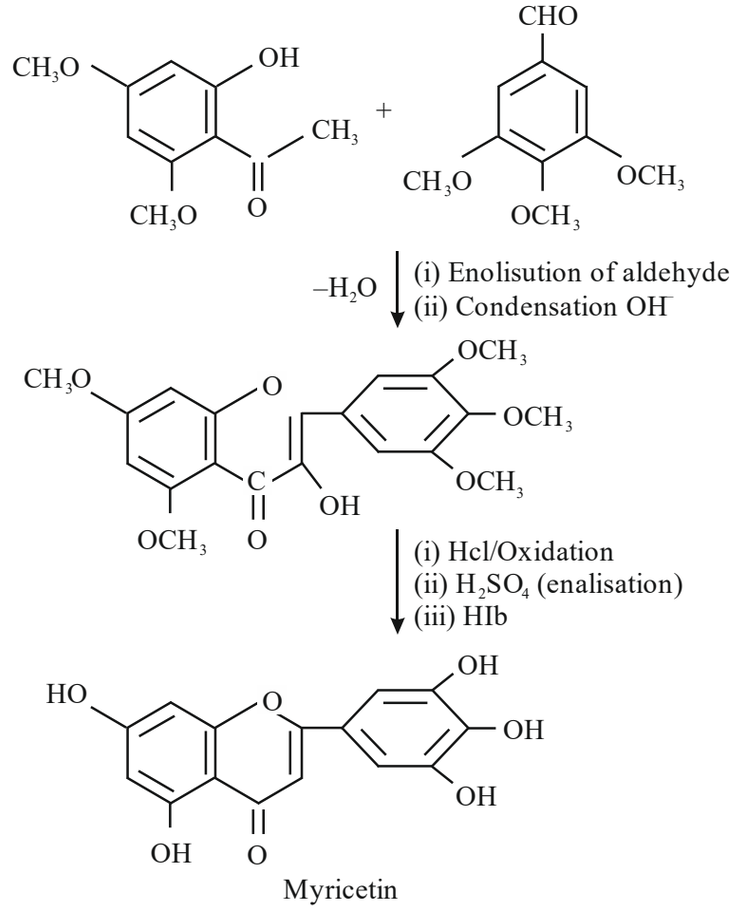
5. **Synthesis of Myricetin (Kostanecki Synthesis)**

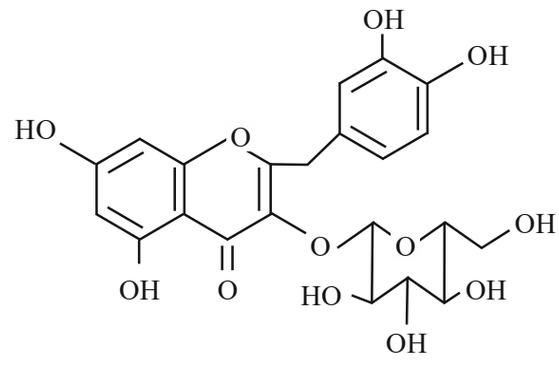


Myricetin

4.2.5 Quercetin 3-Glucoside ($C_{27}H_{20}O_{12}$)

1. **IUPAC Name:** 3-{[(2S, 3R, 4R, 5R)-5-(1R)-1, 2-dihydroxythyl]-3, 4-dihydroxyoxolan-2-yl] oxy}-2-(3, 4-dihydroxyphenyl)-5, 7-dihydroxy-4H-Chromon-4-one.

2. **Structure**



Quercetin-3 Glucoside

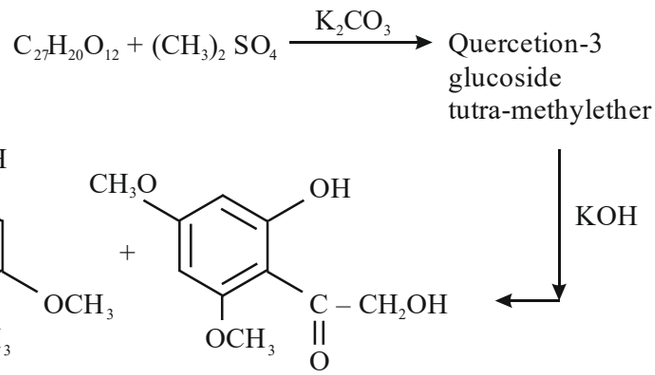
Fig. 4.6 *Structure of Quercetin 3-Glucoside*

Constitution

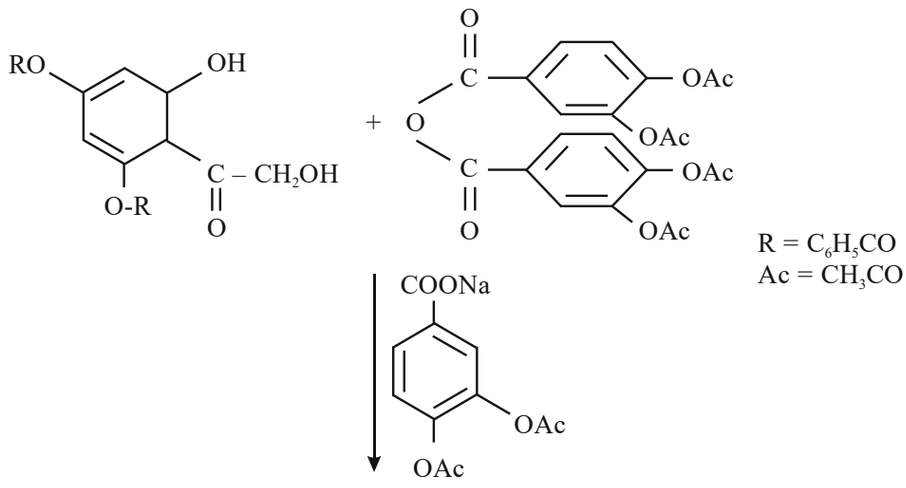
1. Molecular formula of quercetin-3 glucoside is $C_{27}H_{20}O_{12}$
2. On hydrolysis, it forms quercetin and glucose.

$$\underset{\text{Quercetin-3-Glucoside}}{C_{27}H_{20}O_{12}} + H_2O \longrightarrow \underset{\text{Quercetin}}{C_{15}H_{10}O_7} + \underset{\text{Glucose}}{C_6H_{12}O_6}$$

3. **Presence of Sugar Moiety:** In quercetin-3-glucoside, glucose is present as sugar moiety and is confirmed by carbohydrate test.
4. Quercetin-3-glucoside is a common flavonol. Glycoside, is confirmed by appearance of dark brown or black colour in ultraviolet light.
5. Attachment of glucose unit at C_3 is confirmed by Karrer method.
6. On methylation of quercetin-3 glucoside with dimethyl sulphate in presence of potassium carbonate forms tetra methyl ether derivative. When tetramethyl derivative is boiled with ethanolic solution of potassium hydroxide or barium hydroxide it forms w-6 dihydroxy-2, 4-dimethyl acetophenone and veratric acid.



7. Synthesis of quercetin-3-glucoside



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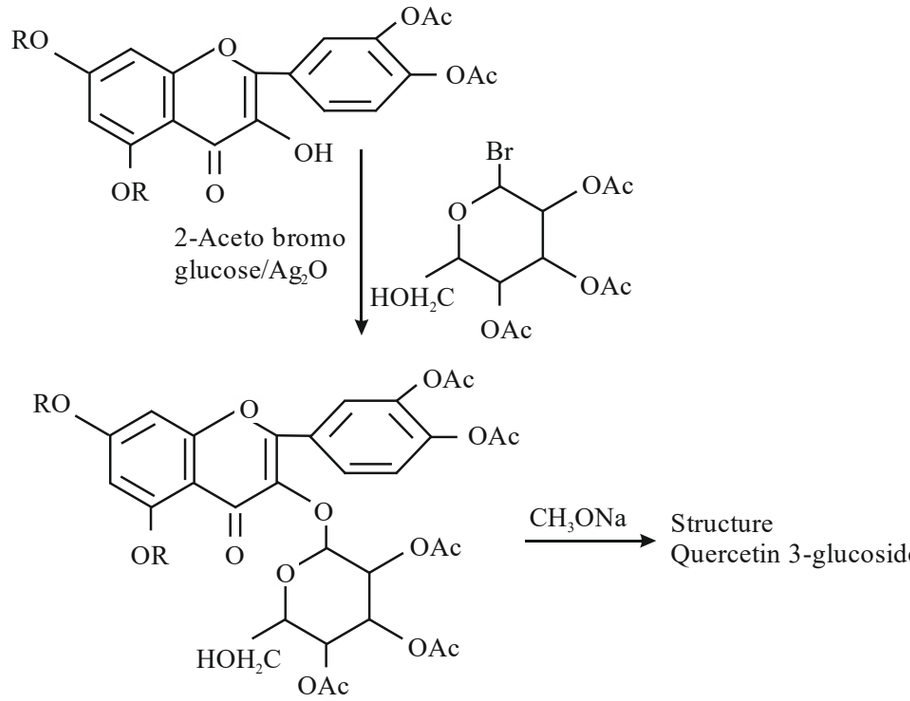
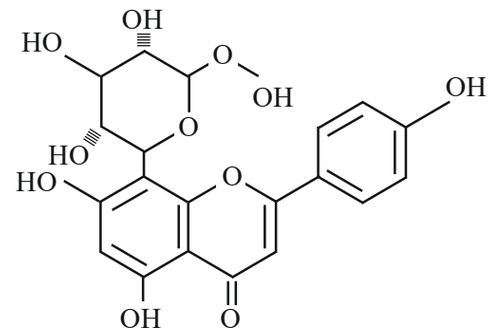


4.2.6 Vitexin ($C_{21}H_{20}O_{10}$)

IUPAC Name: 8-(β-D-Glucosepyranosyl)-4, 5, 7-trihydroxy flavone

Structure

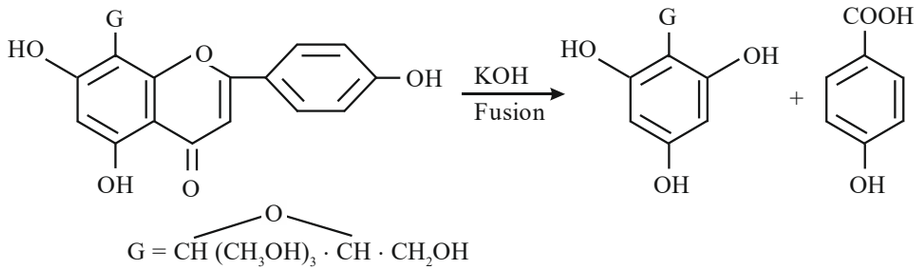


Vitexin

***Fig. 4.7** Structure of Vitexin*

Constitution

1. Molecular formula of vitexin is $C_{21}H_{20}O_{10}$.
2. Its hydrolysis with alkali or acid is difficult which indicate that molecule is having a C-glucosyl derivative.
3. Vitexin when reacts with $FeCl_3$ forms free sugar and apigenin flavonoid.
4. By the oxidation, it gives one mole of formic acid.
5. **Fusion with KOH:** Vitexin on KOH degradation gives p-hydroxy benzoic acid and c-glycosyl derivative of phloroglucinol



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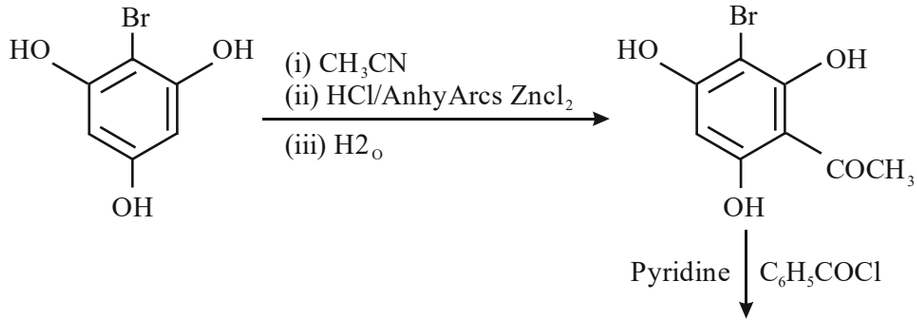
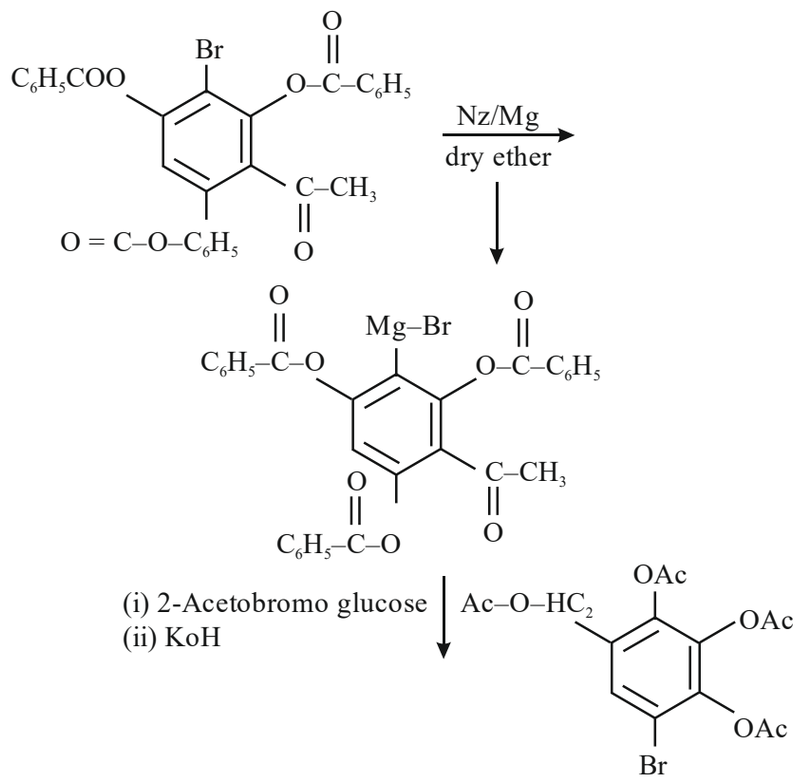
6. **Synthesis of Vitexin:** Structure of vitexin is confirmed by synthesis





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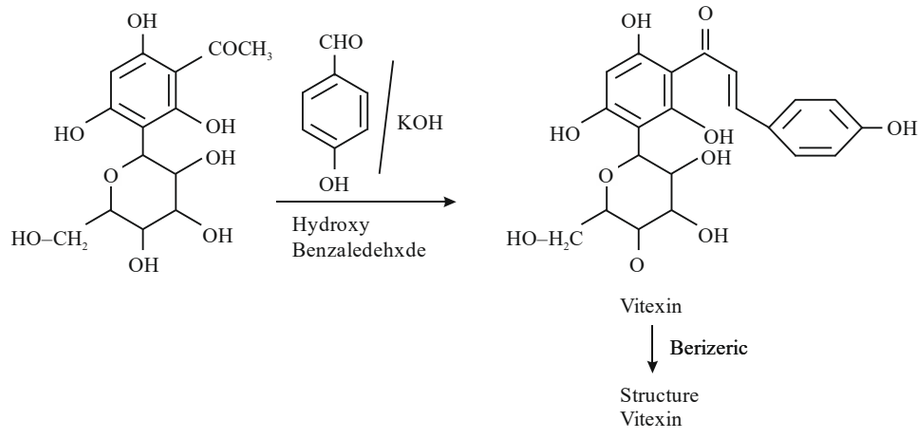


4.2.7 Daidzein ($C_{15}H_{10}O_4$)

Daidzein is an isoflavone pigment. The isoflavones are naturally occurring compounds. Daidzein is found in all vegetables and fruits also.

1. IUPAC Name 7-Hydroxy-3-(4-hydroxyphenyl) chromene-4-one.
2. Structure

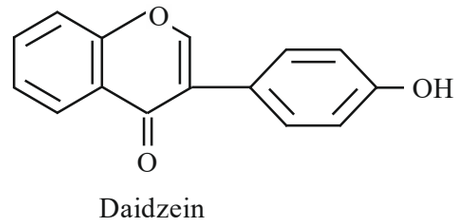


Fig. 4.8 *Structure of Daidzein*

Constitution

(i) **Molecular Formula:** Molecular formula of daidzein is $C_{15}H_{10}O_4$.

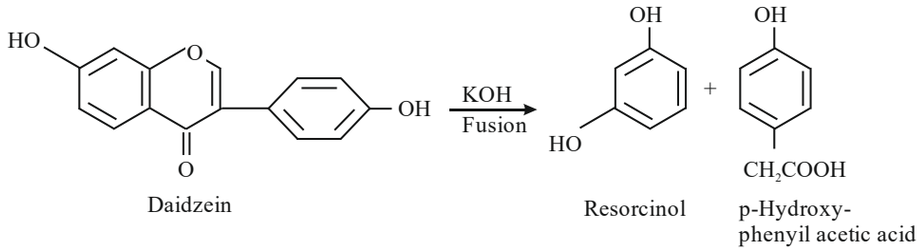
(ii) **Presence of Hydroxyl Group:** Daidzein forms diacetyl derivative on acetylation with acetic di-hydride in the presence of fused sodium acetate.

$$C_{15}H_8O_2(OH)_2 \xrightarrow[CH_3COONa]{2(CH_3CO)_2O} C_{15}H_8O_2(O-\overset{\overset{O}{\|}}{C}-CH_3)_2 + 2CH_3COOH$$

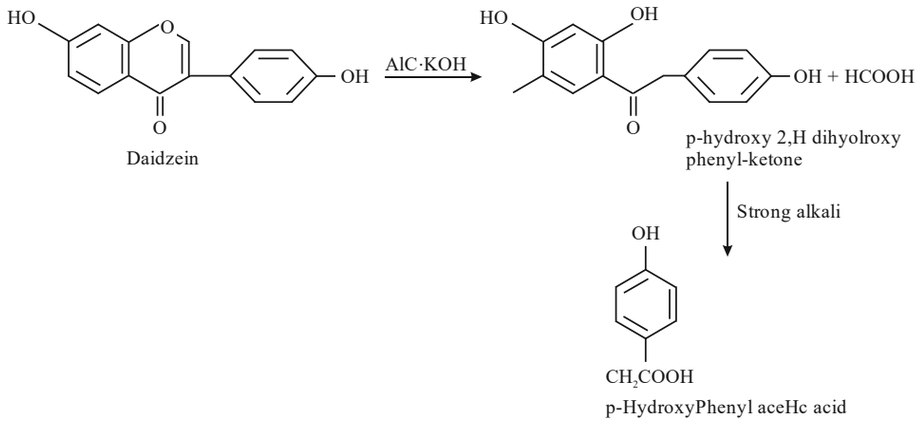
diacetyl diadzein

(iii) On methylation, test shows that daidzein does not contain any methoxy group.

(iv) **Fusion with KOH:** Daidzein fused with KOH gives resorcinol and p-hydroxy phenyl acetic acid.



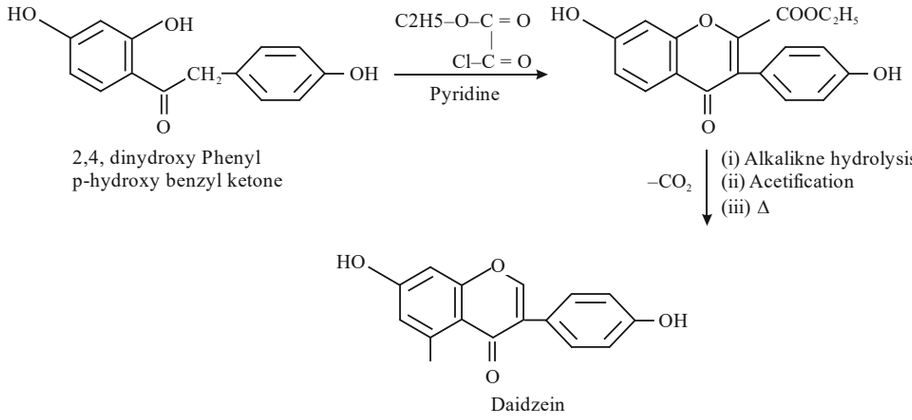
(v) Degradation with alcoholic KOH gives formic acid and p-hydroxy 2, 4 dihydroxyphenyl ketone, further degradation of p-hydroxy 2, 4 dihydroxyphenyl with strong alkali gives p-hydroxy phenyl acetic acid.



The formation of these product could be explained if ring A has hydroxyl group at position VII and ring B has hydroxyl group at position-IV.

3. **Synthesis of Diadzein:** Diadzein structures confirmed by following synthesis methods

   (i) Baker-Ollis method (1953)



4.2.8 Butein

Molecular Formula: $C_{15}H_{12}O_5$ **Molecular Weight:** 272.25

IUPAC name: 22 ,3,4,42 -Tetrahydroxychalcone

Butein is a flavonoid obtained from the seed of Cyclopia subternata. It is a specific protein tyrosine kinase inhibitor that induces apoptosis.

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Butein is a chalcone that is (E)-chalcone bearing four additional hydroxy substituents at positions 2', 3, 4 and 4'. It has a role as a tyrosine kinase inhibitor, an antioxidant, an EC 1.1.1.21 (aldehyde reductase) inhibitor, an antineoplastic agent, a geroprotector, a radiosensitizing agent, a hypoglycemic agent and a plant metabolite.

It is a member of chalcones and a polyphenol. It has antioxidative, aldose reductase and advanced glycation endproducts inhibitory effects. It is also a sirtuin-activating compound, a chemical compound having an effect on sirtuins, a group of enzymes that use NAD+ to remove acetyl groups from proteins.

Buteins possess a high ability to inhibit aromatase process in the human body, for this reason, the use of these compounds in the treatment of breast cancer on the estrogen ground has been explored.

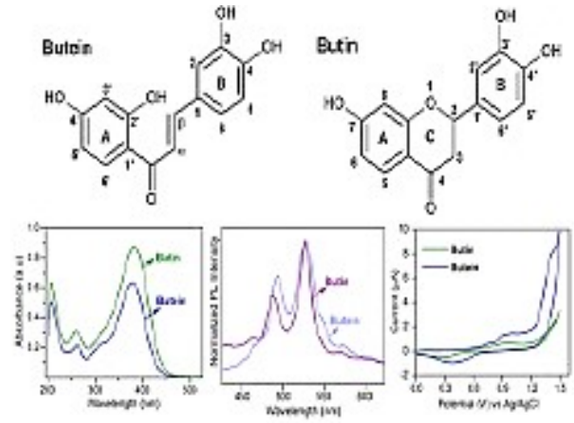


Fig. 4.9 *Structure of Butein*

4.2.9 Aureusin

Molecular Formula: $C_{21}H_{20}O_{11}$

Molecular Weight: 448.4

IUPAC Name: 2-[(3,4-dihydroxyphenyl)methylidene]-4-hydroxy-6-[(2S,4S,5S)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy-1-benzofuran-3-one.

Aureusin is a member of the class of compounds known as aurone o-glycosides. Aurone o-glycosides are aurone flavonoids containing a carbohydrate moiety O-glycosidically bound to the aurone skeleton. Aureusin is slightly soluble (in water) and a very weakly acidic compound (based on its pKa). Aureusin can be found in lemon, which makes aureusin a potential biomarker for the consumption of this food product.

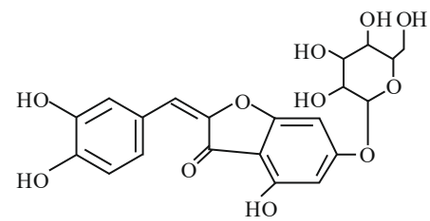


Fig. 4.10 *Structure of Aureusin*

4.2.10 Cyanidin-7-Arabinoside ($C_{20}H_{19}O_{10}$)

Cyanidin 7-arabinoside is found in pines. It is isolated from the apple tree.

1. IUPAC Name: 2-[2-(3, 4 dihydroxyphenyl)-3, 5-dihydroxychromenylium -7-yl] oxyoxane 3-45-trial.
2. Structure

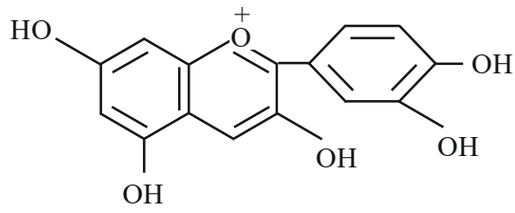


Fig. 4.11 *Structure of Cyanidin-7-arabinoside*

Constitution

(i) Molecular formula of Cynidin 7-arabinoside is $C_{20}H_{19}O_{10}$.

(ii) Cyanidin 7-arabinoside on hydrolysis with hydrochloric acid forms cyanidin and arabinose

$$\underset{\text{Cyanidine derivatives}}{C_{20}H_{19}ClO_{10}} + H_2O \xrightarrow{HCl} \underset{\text{Cyanidin}}{C_{15}H_{11}ClO_6} + \underset{\text{Arabinose}}{C_5H_{10}C_5}$$

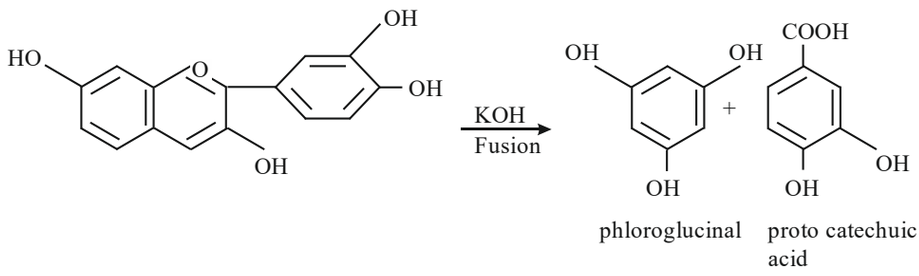
(iii) On acetylation, cyanidin gives penta acetyl cynaidin. It shows that 5 hydroxyl group are present.

$$\underset{\text{Cyanidine Chloride}}{C_{15}H_6ClO(OH)} + 5(CH_3CO)_2O \xrightarrow{CH_3COONa}$$

$$\underset{\text{Penta acetyl derivative cyanidin chloride}}{C_{15}H_6ClO(OCOCH_3)_5} + 5CH_3COOH$$

(iv) On Methylation, test shows that cyanidin chloride does not contain any methoxy group.

(v) **Fusion with KOH:** Cyanidin chloride fused with KOH gives phloroglucinol and protocatechuic acid



3. **Synthesis:** The structure of cyanidin chloride has been confirmed by its synthesis given by Robinson (1928).

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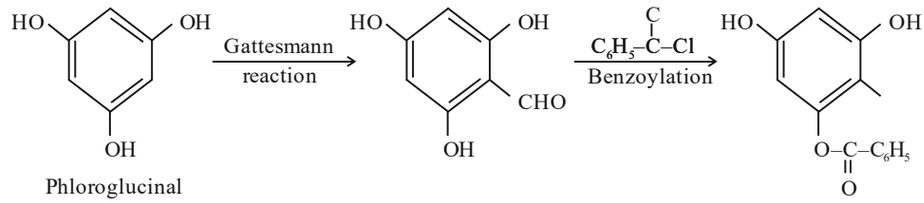
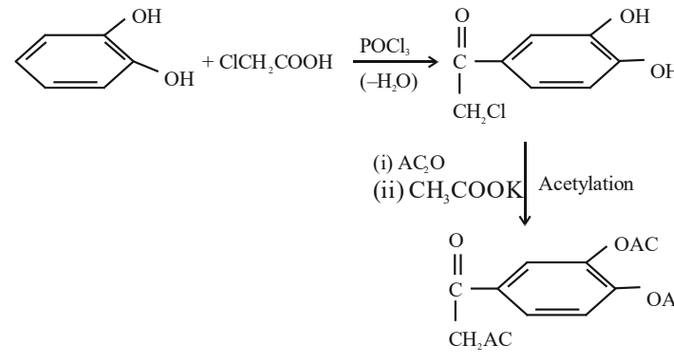
The various steps of this synthesis are as follows:

(A) Synthesis of 2-Benzoylphloroglucinaldehyde



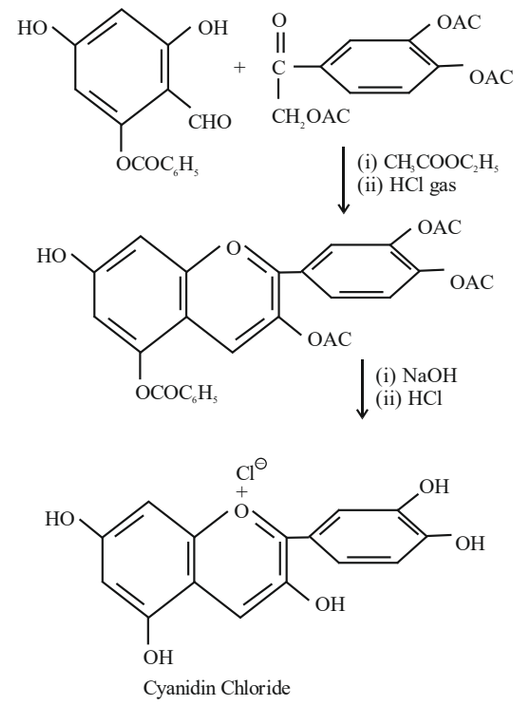
Phloroglucinal

(B) Synthesis of w, 3, 4-triacetoxyacetophenone



ω, 3, 4-triacetoxy
acetoxyacetophenene

(C) Synthesis of cyanidin chloride by reaction of 2 benzoyloxy-4, 6 dihydroxy benzaldehyde and w, 3 triacetoxy acetoxyacetophenone.

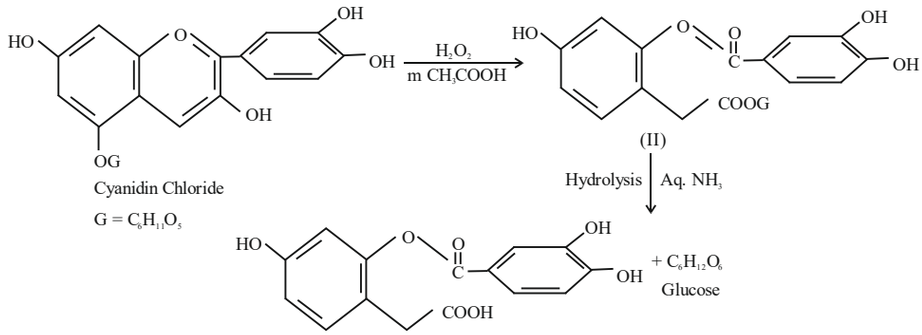


Cyanidin Chloride

4. Position of Glucose unit in cyanine molecule

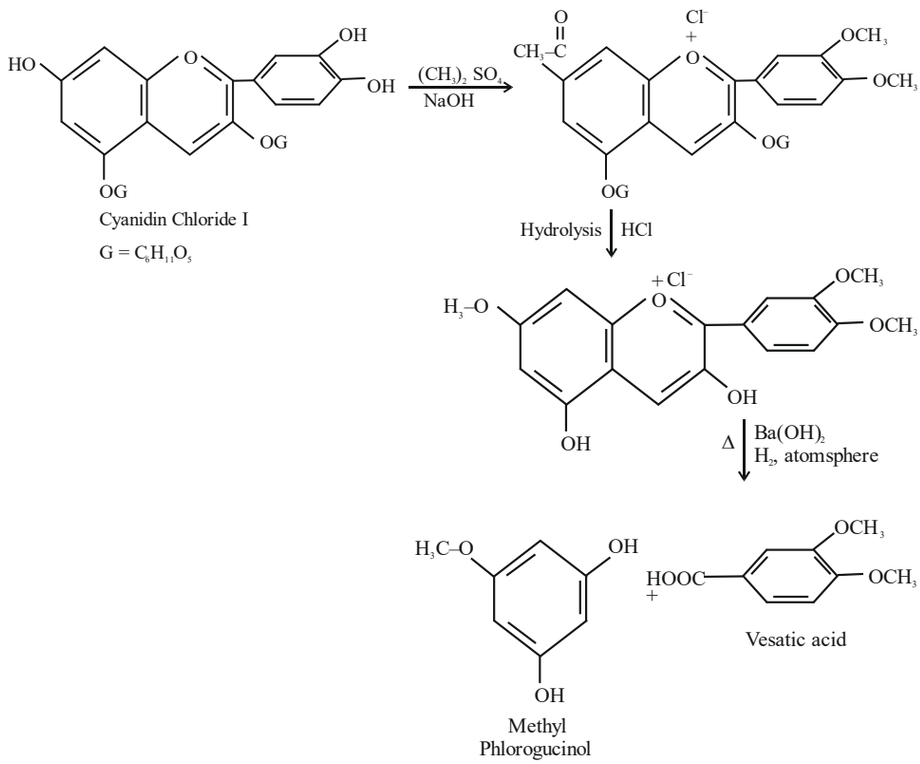
(i) Position of One Glucose Unit

(a) **Karrer's Method (1927):** Cyanin chloride is treated with 15% hydrogen peroxide in acetic acid which opens the heterocyclic ring by breaking C_2–C_3 bond, without removing the sugar residue. The latter mole compound when treated with aqueous ammonia, undergoes hydrolysis and one molecule of glucose is easily removed.



(ii) Position of Second Glucose Unit

On methylation of cyanin chloride, it gives tri-methyl derivative. The methylated product is hydrolysed with HCl to remove sugar residue and then with $Ba(OH)_2$, yields methyl pholoroglucinol and veratic acid. The two free hydroxyl groups in mono-methyl phloroglucinol reveals the position of sugar residue indicates that other glucose residue is present in position 5.



NOTES

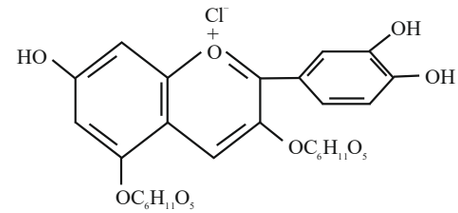
Finally, the synthesis of cyanin clearly establishes the glucose residue in the position 3 and 5.

NOTES

4.2.11 Cyanidin ($C_{15}H_{11}O_6Cl$)

Cyanidin was fist anthocyanin which was obtained in the form of its crystalline chloride. It is isolated from red rose and conflowers.

Structure



Cyanidin Chloride

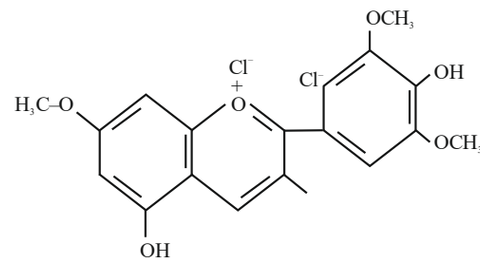
Isolation and synthesis: Write the synthesis of Cyanidin-7-Arabinoside from page 115-116

4.2.12 Hirsutidin Chloride ($C_{30}H_{37}ClO_{17}$)

Hirsutidin is found in primulahirsuta. It is obtained by the hydrolysis with HCl.

IUPAC Name: 3, 4′, 5 trihydroxy 3′, 5′, 7 trimethoxy flavylium chloride.

Structure



Hissutidin Chloride

Fig. 4.12 *Structure of Hirsutidin Chloride*

Constitution

(1) **Molecular formula:** The molecular formula of hirsutidin chloride is $C_{30}H_{37}ClO_{17}$.

(2) **Presence of Hydroxyl Group**

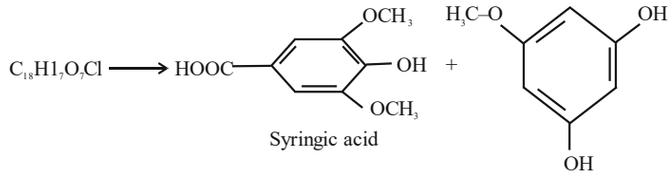
(i) Hirsutidin chloride forms triacetoxy derivation on acetylation with acetic anhydride in the presence of fused potassium acetate

$$\underset{\text{Hirsutidin Chloride}}{C_{18}H_{17}ClO_4(OH)_3} \xrightarrow[CH_3COOK]{(CH_3CO)_2O} \underset{\text{Trihydroxy derivative of Hirsutidin Chloride}}{C_{18}H_{13}ClO_4(O\text{–}\overset{O}{\overset{\|}{C}}\text{–}CH_3)_3}$$

(ii) On methylation, hirsutidin chloride shows three methoxy groups present in the molecule.

(iii) Hirsutidin chloride shows peak at 17-20 cm^{-1} in infrared spectrum.

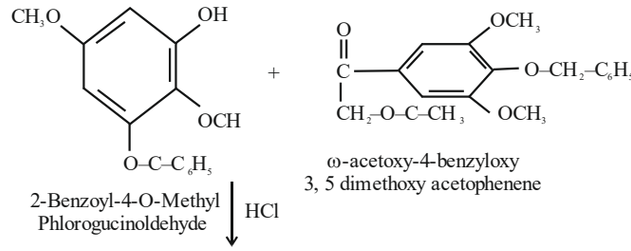
(iv) By the degradation method, hirsudition chloride is boiled with $Ba(OH)_2$ solution in presence of hydrogen atmosphere it gives syringic acid and monomethyl phloroglucinol.



Syringic acid

Monomethyl Phloroglucinol

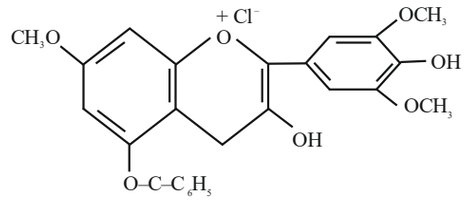
3. Synthesis (Robinson and his Co-workers 1930)



2-Benzoyl-4-O-Methyl Phloroglucinoldehyde

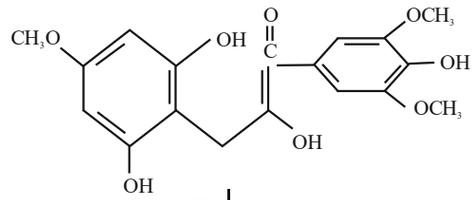
ω-acetoxy-4-benzyloxy 3, 5 dimethoxy acetophenene

HCl



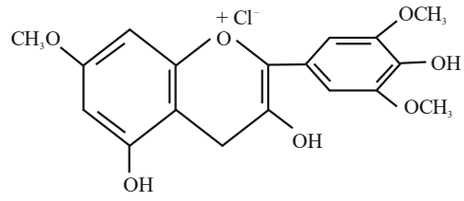
N_2 atmosphere

NaOH



N_2 atmosphere

HCl



Hirsutidin Chloride

NOTES

NOTES

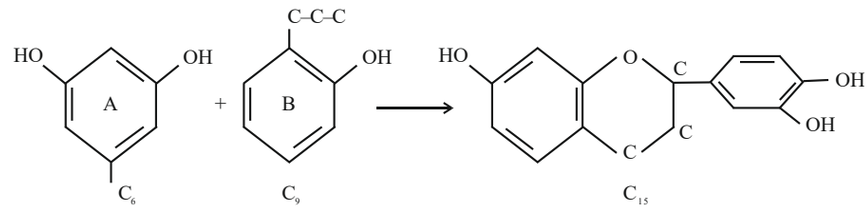
Check Your Progress

1. What do you understand by apigenin?
2. What is the molecular formula of apigenin?
3. Give the IUPAC name of leutolin.
4. Write the molecular formula of quercetin.
5. What is the absorption maxima of myricetin?
6. Write the IUPAC name of quercetin-3-glucoside.
7. Which product is formed when vitexin reacts with FeCl3 (write 3 in subscript)?
8. What is daidzein?
9. Give the molecular formula of cyanidin.
10. At what wavelength hirsutidin chloride shows maximum peak in an IR spectrum?

4.3 BIOYNTHESIS OF FLAVONOIDS

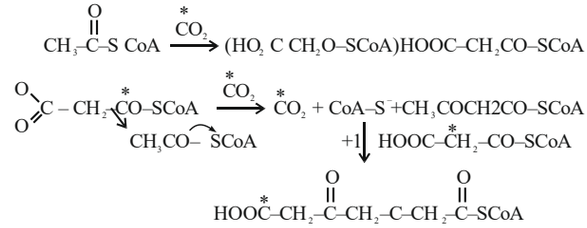
Synthesis of flavonoids was given by Robinson in 1936 considered that the flavonoids skeleton is made by C_{15} and it is divided in two parts C_6 and C_9.

In the biosynthesis process of flavonoids we know that ring A and B are formed by the different routes. Ring A is formed by acetate pathway and ring B is formed by Shikimic pathway.



4.3.1 Acetate Pathway

It was proposed by Birch by the synthesis of fatty acid, it has been assumed that malonyl coenzyme A than acetyl coenzyme A is the intermediate in flavonoid biosynthesis. By Lynen and his co-workers, feeding experiment in the study of biosynthesis of fatty acid shows that malonate was an excellent precursor. Experiments using labelled sodium hydrogen carbonate ($NaHCO_3$) showed that this was not incorporated with labelled acetate. Therefore a possible route for biosynthesis of C_6– is



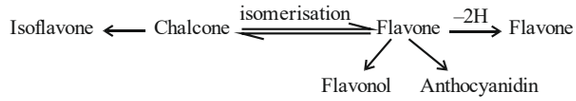
4.3.2 Shikimic Acid Pathway

C_6 and C_3 unit arise this pathway.

The route conversion of shikmic acid to cimmamic acid indicate that they are good precursors for quercetin.

NOTES

The co-occurrence of anthocyanidin and flavonal, flavone may mean a step in pigment synthesis. The possible sequence appears to be.



Check Your Progress

11. Who gave the biosynthesis of flavonoids?
12. Flavonoids skeleton is divided into how many parts?
13. How many rings are there in a flavonoid biosynthesis?
14. Which units arise from shikimic acid pathway?

4.4 ANSWERS TO 'CHECK YOUR PROGRESS'

1. Apigenin is a trihydroxyflavone that is flavone substituted by hydroxy groups at positions 4', 5 and 7.
2. The molecular formula of apigenin is $C_{15}H_{10}O_5$.
3. IUPAC name of leutolin is 5-7-dihydroxy-2-(3, 4-dihydroxy phenyl)-chromene-4-one.
4. Molecular formula of quercetin is $C_{15}H_{10}O_7$.
5. Absorption maxima λ_{max} of myricetin is at 350-390 nm (band II) and 250-270 nm (band I) in UV-spectrum.
6. IUPAC name of quercetin-3-glucoside is 3-{[(2S, 3R, 4R, 5R)-5-(1R)-1, 2-dihydroxythyl]-3, 4-dihydroxyoxolan-2-yl] oxy}-2-(3, 4-dihydroxyphenyl)-5, 7-dihydroxy-4H-Chromon-4-one.
7. Vitexin when reacts with $FeCl_3$ forms free sugar and apigenin flavonoid.
8. Daidzein is an isoflavone pigment, found in all vegetables and fruits.
9. Molecular formula of cyaniding is $C_{15}H_{11}O_6Cl$.
10. Hirsutidin chloride shows peak at 17-20 cm–1 in infrared spectrum.
11. Synthesis of flavonoids was given by Robinson in 1936.
12. A flavonoids skeleton is made by C_{15} and it is divided in two parts C_6 and C_9.
13. There are two rings, Ring A and Ring B formed in a flavonoid biosynthesis.
14. C_6 and C_3 unit arise in shikimic acid pathway.

4.5 SUMMARY

NOTES

- Plant pigments or phytochromes are the substances produced by plants that have a colour resulting from selective colour absorption.
- There are many different plant pigments, and they belong to different classes of organic compounds.
- Pigments play an important role in the pollination as well where pigment accumulation or loss can lead to change in the floral colour, signalling to pollinators which flowers are rewarding and contain more pollen and nectar.
- There are different type of pigments that a plant can have. The variation in plant pigments are due to the different wavelength of light they absorb.
- Plant pigments are important in controlling photosynthesis, growth and development of plants.
- Pigments also protect plant from damage caused by UV and visible light.
- Apigenin is a trihydroxyflavone that is flavone substituted by hydroxy groups at positions 4', 5 and 7.
- The molecular formula of apigenin is $C_{15}H_{10}O_5$.
- It shows absorption maxima λ_{max} at 267nm (bond II) and 336V nm (bond I) in UV-spectrum.
- Tri-methoxy apigenin on oxidation with $KMnO_4$ gives p-methoxybenzoic acid as one of the products.
- Apigenin on KOH degradation gives phloroglucinol and p-Hydroxy benzoic acid.
- Luteolin is a tetrahydroxyflavone in which the four hydroxy groups are located at positions 3', 4', 5 and 7.
- Its IUPAC name is 5-7-dihydroxy-2-(3, 4-dihydroxy phenyl)-chromene-4-one.
- Molecular formula of luteolin is $C_{15}H_{10}O_6$
- Alcoholic solution of luteolin gives dark pink colour with Mg turnings and dilute HCl.
- Luteolin on acetylation with acetic anhydride and fused sodium acetate yields tetra-acetyl luteolin, indicating the presence of four hydroxyl groups.
- By the process of KOH degradation, luteolin gives phloroglucinol and dihydroxy benzoic acid.
- Quercetin is a plant pigment (flavonoid) found in many plants and foods, such as, onions, green tea, apples, and berries.
- Its IUPAC name is 2-(3, 4-dihydroxy phenyl) -3, 5, 7 trihydroxy chromen-4-one.
- Molecular formula of quercetin is $C_{15}H_{10}O_7$.
- By the process of KOH degradation, quercetin gives phloroglucinal phloroglucinol and protocatechuic acid.

NOTES

- Myricetin is a hexahydroxyflavone that is flavone substituted by hydroxy groups at positions 3, 3', 4', 5, 5' and 7.
- Its IUPAC name is 3, 5, 7-trihydroxy-2- (3, 4, 5-trihydroxyphenyl)-4 H-1-benzopyram-4-one.
- On acetylation, myricetin gives hexa-acetate derivative. Myricetin on methylation gives hexamethyl myricetin.
- It shows absorption maxima λ_{max} at 350-390 nm (band II) and 250-270 nm (band I) in UV-spectrum.
- Alcoholic solution of myricetin gives magenta colour with magnesium and dilute HCl.
- On acetylation, myricetin gives hexa-acetate derivative. Myricetin on methylation gives hexamethyl myricetin.
- Myricetin on KOH degradation gives phloroglucinol and gallic acid.
- Molecular formula of quercetin-3 glucoside is $C_{27}H_{20}O_{12}$.
- IUPAC Name of Quercetin 3-Glucoside is 3-{[(2S, 3R, 4R, 5R)-5-(1R)-1, 2-dihydroxythyl]-3, 4-dihydroxyoxolan-2-yl] oxy}-2-(3, 4-dihydroxyphenyl)-5, 7-dihydroxy-4H-Chromon-4-one.
- Molecular formula of quercetin-3 glucoside is $C_{27}H_{20}O_{12}$
- In quercetin-3-glucoside, glucose is present as sugar moiety and is confirmed by carbohydrate test.
- On methylation of quercetin-3 glucoside with dimethyl sulphate in presence of potassium carbonate forms tetra methyl ether derivative.
- Vitexin is an apigenin flavone glycoside, which is found in the passion flower, bamboo leaves and pearl millet.
- Its IUPAC name is 8-(β-D-Glucosepyranosyl)-4, 5, 7-trihydroxy flavone.
- Molecular formula of vitexin is $C_{21}H_{20}O_{10}$.
- Vitexin when reacts with $FeCl_3$ forms free sugar and apigenin flavonoid.
- Daidzein is an isoflavone pigment.
- Daidzein is found in all vegetables and fruits also.
- Its IUPAC name is 7-Hydroxy-3-(4-hydroxyphenyl) chromene-4-one.
- Molecular formula of daidzein is $C_{15}H_{10}O_4$.
- Daidzein forms diacetyl derivative on acetylation with acetic di-hydride in the presence of fused sodium acetate.
- Daidzein fused with KOH gives resorcinol and p-hydroxy phenyl acetic acid.
- Cyanidin 7-arabinoside is found in pines. It is isolated from the apple tree.
- Molecular formula of Cynidin 7-arabinoside is $C_{20}H_{19}O_{10}$.
- On acetylation, cyanidin gives penta acetyl cynaidin. It shows that 5 hydroxyl group are present.
- Cyanidin chloride fused with KOH gives phloroglucinol and protocatechuic acid.

NOTES

- Cyanidin was fist anthocyanin which was obtained in the form of its crystalline chloride. It is isolated from red rose and conflowers.
- Hirsutidin is found in primulahirsuta. It is obtained by the hydrolysis with HCl.
- The molecular formula of hirsutidin chloride is $C_{30}H_{37}ClO_{17}$.
- Hirsutidin chloride forms triacetoxy derivation on acetylation with acetic anhydride in the presence of fused potassium acetate.
- Hirsutidin chloride show pick at 17-20 cm^{-1} in infrared spectrum.
- Synthesis of flavonoids by Robinson in 1936 considered that the flavonoids skeleton is made by C_{15} and it is divided in two parts C_6 and C_9.
- In the biosynthesis process of flavonoids we know that ring A and B are formed by the different routes. Ring A is formed by acetate pathway and ring B is formed by Shikimic pathway.

4.6 KEY TERMS

- **Plant Pigments:** Plant pigments or phytochromes are the substances produced by plants that have a colour resulting from selective colour absorption.
- **Apigenin:** Apigenin is a trihydroxyflavone that is flavone substituted by hydroxy groups at positions 5 and 7.
- **Leutolin:** Luteolin is a tetrahydroxyflavone in which the four hydroxy groups are located at positions 3', 4', 5 and 7.
- **Quercetin:** Quercetin is a plant pigment (flavonoid) found in many plants and foods, such as, onions, green tea, apples, and berries.
- **Myricetin:** Myricetin is a hexahydroxyflavone that is flavone substituted by hydroxy groups at positions 3, 3', 4', 5, 5' and 7.
- **Vitexin:** Vitexin is an apigenin flavone glycoside, which is found in the passion flower, bamboo leaves and pearl millet.
- **Daidzein:** Daidzein is an isoflavone pigment, found in all vegetables and fruits.
- **Cyanidin:** Cyanidin 7-arabinoside is found in pines. It is isolated from the apple tree.

4.7 SELF ASSESSMENT QUESTIONS AND EXERCISES

Short Answer Questions

1. What do you understand by apigenin? How it is synthesized?
2. State the position of hydroxyl group in leutolin.
3. State the synthesis quercetin.
4. What is the constitution of quercetin-3-glycoside?

5. What is the acetate pathway of biosynthesis of flavonoids?
6. What is the Shikimic acid pathway of flavonoid biosynthesis?

Long Answer Questions

1. Discuss in detail the position of glucose in cyanine molecule.
2. Discuss the synthesis of vitexin.
3. Explain the constitution and synthesis of myricetin.
4. Explain the isolation and synthesis of cyanidin.
5. What do you understand by flavonoids? Describe in detail the process of biosynthesis of flavonoids.

NOTES

4.8 FURTHER READING

Krishnaswamy, N. R. 1999. *Chemistry of Natural Products A Unified Approach*. Himayatnagar, Hyderabad: Universities Press.

Bhat, Sujata V., B.A. Nagasampagi, Meenakshi Sivakumar · 2005. *Chemistry of Natural Products*. Narosa Publishing House

Rahman,Atta-ur. 2020. *Studies in Natural Products Chemistry.* Volume 67. Amsterdam, Netherlands: Elsevier Science.

Cooper, Raymond. , George Nicola. 2014. *Natural Products Chemistry-Sources, Separations and Structures.* London UK: Taylor & Francis.

UNIT 5 PORPHYRINS PROSTAGLANDINS PYRETHROIDS AND ROTENONE

Structure

5.0 INTRODUCTION

Porphyrins are the conjugate acids of ligands that bind metals to form complexes. Porphyrins are essential for the function of haemoglobin and protein in our red blood cells that links to porphyrin, binds iron, and carries oxygen to your organs and tissues. Hemoglobin or Haemoglobin, (Hb or Hgb), is the iron-containing oxygen-transport metalloprotein in the red blood cells of almost all vertebrates as well as the tissues of some invertebrates. Haemoglobin in blood carries oxygen from the lungs or gills to the rest of the body. Chlorophyll is one of the most important class of pigments involved in photosynthesis, the process by which light energy is converted to chemical energy through the synthesis of organic compounds.

The prostaglandins are a group of lipids made at sites of tissue damage or infection that are involved in dealing with injury and illness. They control processes, such as inflammation, blood flow, and the formation of blood clots and the induction of labour.

Pyrethroids are a group of man-made pesticides similar to the natural pesticide pyrethrum, which is produced by chrysanthemum flowers. Pyrethroids are used as commercial and household insecticides. Whereas, the rotenone is an odorless, colourless, crystalline isoflavone used as a broad-spectrum insecticide, piscicide, and pesticide. It occurs naturally in the seeds and stems of several plants, such as the jicama vine plant, and the roots of several members of Fabaceae.

In this unit, you will study about the porphyrins, structure and synthesis of haemoglobin and chlorophyll, nomenclature and classification of prostaglandins, synthesis and reactions of pyrethroids and rotenone.

5.1 OBJECTIVES

- Understand the porphyrins
- Illustrate the structure and synthesis of haemoglobin and chlorophyll
- Know about the nomenclature and classification of prostaglandins
- Analyse the synthesis and reactions of pyrethroids and rotenone

5.2 PROPHYRINS

Porphyrins are a group of heterocyclic, macrocycle ***organic compounds***, composed of four modified ***pyrrole subunits*** which contain interconnected at their α-carbon atoms via '***Methine Bridges'* (=CH–).** The parent of porphyrin is porphine, a rare chemical compound of exclusively theoretical interest. Substituted porphines are called *porphyrins*. With a total of 26 π-electrons, of which 18 π-electrons form a planar, continuous cycle, the *porphyrin ring* structure is often described as aromatic. The 18-electron cycle of porphin, the parent structure of porphyrin, (Refer Figure 5.1). One result of the large conjugated system is that porphyrins typically absorb strongly in the visible region of the *electromagnetic spectrum*, i.e., they are deeply coloured. The name 'Porphyrin' derives from the Greek word *porphyra,* which mean **purple**.

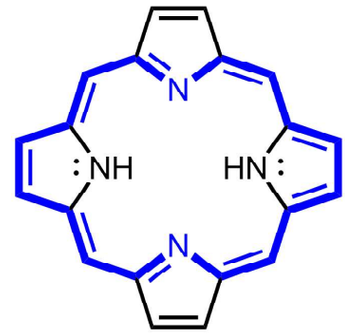


Fig. 5.1 *18-Electron Cycle of Porphin*

Metal complexes derived from porphyrins occur naturally. One of the best-known families of porphyrin complexes is ***heme***, the pigment in red blood cells, a cofactor of the protein ***haemoglobin***.

A porphyrin without a metal-ion in its cavity is a *free base*. Some iron-containing porphyrins are called *hemes*. Heme-containing proteins, or haemoproteins, are found extensively in nature. ***Haemoglobin*** and ***myoglobin*** are two O_2-binding proteins that contain iron porphyrins. Various cytochromes are also hemoproteins.

Heme Porphyrin

While the haemoglobin and myoglobin molecules are very large, complex proteins, the active site is actually a non-protein group called *heme*. The heme consists of a flat organic ring surrounding an iron atom. The organic part is a ***porphyrin ring***

based on porphin (a tetrapyrrole ring), and is the basis of a number of other important biological molecules, such as, chlorophyll and cytochrome. The ring contains a large number of conjugated double bonds, which allows the molecule to absorb light in the visible part of the spectrum. The iron atom and the attached protein chain modify the wavelength of the absorption and gives haemoglobin its characteristic colour. Oxygenated haemoglobin (found in blood from arteries) is bright red, but without oxygen present (as in blood from veins), haemoglobin turns a darker red. Venous blood is often depicted as blue in colour in medical diagrams, and veins sometimes look blue when seen through the skin. The appearance of blood as dark blue is a wavelength phenomenon of light, having to do with the reflection of blue light away from the outside of venous tissue if the vein is ~0.02 inches deep or more.

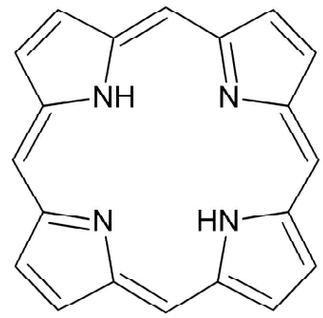


Fig. 5.2 *Porphin - Building Block of Heme*

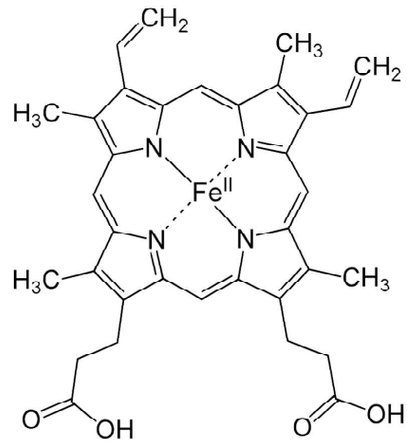


Fig. 5.3 *Heme, Non-Protein Active Site within Myoglobin and Haemoglobin*

The *porphyrins* are heterocyclic ring structures that include four pyrrole rings joined together through carbon (methenyl) bridges. The most abundant porphyrins in nature are found in haemoglobin and the chlorophylls. In the centre of porphyrins a metal atom is chelated to the nitrogen atoms of the pyrrole units.

Chlorophyll is the green pigment in plants, algae, and cyanobacteria, i.e., essential for *photosynthesis*. Its central structure is an aromatic porphyrin or chlorine (reduced porphyrin) ring system with a sequestered magnesium atom. A fifth ring is fused to the porphyrin. The porphyrins have been selected by the fine comb of evolution as the functional pigment of biology. As heme they are part of the electron

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transport system of almost all cells. As chlorophyll they are at the heart of photosynthesis. They are involved in the critical energy conversion step of photosynthesis.

5.2.1 Information about Haemoglobin

Definition

Haemoglobin is a conjugated chromoprotein which contains haeme as prosthetic group and globin as the apoprotein part. Conjugated proteins are proteins which have a protein part known as apoprotein and a non-protein part known as prosthetic group. Chromoproteins are proteins which are coloured.

General Information About Hb

In adult human beings, the normal concentration of haemoglobin present is 13–18 gm/dl. There is about 750 g of Hb in the total circulating blood of a 70-kg man. One sub-unit of haemoglobin binds to one molecule of oxygen and one Hb has four subunits. 1 g of Hb can carry 1.34 ml of oxygen. Space occupied by Hb in an erythrocyte is about 35 per cent and contributes to about 97 per cent of dry weight. About 27–32 pg of Hb is present in one molecule of erythrocyte. Approximately 6.25 g of Hb is formed and destroyed in the body each day.

History of Discovery of Hb

Different aspects of haemoglobin (Hb) were discovered at different times. In 1940, Hunefeld discovered the Hb in blood. In 1951, Otto Funko was able to isolate Hb crystals by successively diluting RBCs with a solvent such as pure water, alcohol or ether, followed by slow evaporation of the solvent from the resulting protein solution. Hb's reversible oxygenation was found out by Felix Hoppe–Seyler. Its molecular structure was discovered by Max Perutz by x-ray crystallography in 1959. He received Nobel Prize in chemistry in 1962 along with John Kendrew. Functions of Hb were found out by physiologist Claude Bernard.

Structure of Haemoglobin

Haemoglobin has two parts. They are:

1. Haeme
2. Globin

Structure of Haeme

Haeme is iron-containing porphyrin nucleus. The porphyrins are complex in nature, having a 'Tetra-Pyrrole' (four pyrrole rings) structure which are linked by –CH = bridges called 'Methynyl' or 'Methylidine' bridges. The structure of haeme is shown in Figure 5.4. The valency of iron (Fe) in Hb is six. Iron ion is situated at the centre linking to four nitrogens of the pyrrole rings. The fifth linkage is with the nitrogen of the imidazole ring of Histidine (His) of polypepetide chains, i.e., 'Haeme-Linked' group. In case of α-chain, histidine is located in amino acid number 87 and in case of β-chain, it is located in His 92. The sixth valence is linked to H_2O in deoxygenated Hb. In case of oxygenated Hb, the H_2O is displaced by O_2.

$$Hb.H_2O + O_2 = Hb.O_2 + H_2O$$

Structure of Globin

The protein part of the Hb is globin. Globin has four peptide chains as shown in Figure 5.4. Each α-chain consists of 141 amino acids and each β-chain consists of 146 amino acids. The protein portion of each of this chain is called globin. Each subunit or globin has a molecular weight of about 17,000 dalton which makes the total molecular weight of the tetramer of about 68,000 daltons.

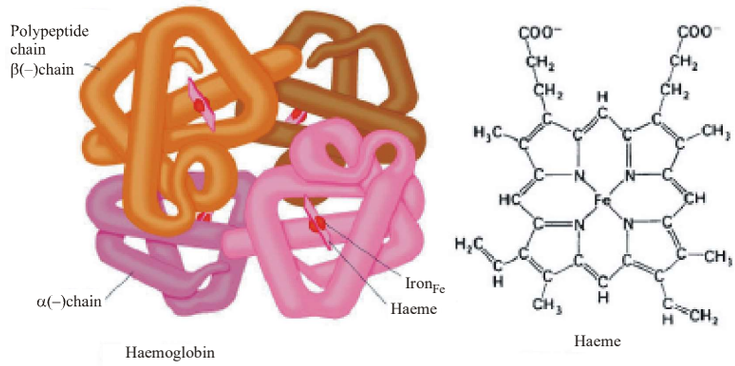


Fig. 5.4 *Structure of Haemoglobin*

Both types of chains are similar in structure. The secondary structure of these four chains is α-helix. Each α-chain and β-chain folds into 8 α-helical segments which further folds to form globular tertiary structure. These four polypeptide chains are arranged in the form of tetrahedron. The four polypeptide chains are then bound to each other by salt bridges, hydrogen bonds and hydrophobic interaction. The folded tertiary structures of helices form pocket. These pockets are known as 'haeme pockets.' In each of these four 'haeme pockets,' one haeme is located. Therefore, each Hb molecule contains four 'haeme' units. The gene located for α-chain is situated on chromosome number 16 and the β, γ and δ-chains are located on chromosome number 11.

Types of Haemoglobin

Haemoglobins can be classified mainly into two types. Some Hbs are normal types and some are abnormal variants.

Normal Haemoglobin

There are a variety of normal human haemoglobins which are made up of four sub-units of polypeptides. The polypeptide sub-units are made of different combinations of five different polypeptide chains. The five sub-units are usually known as α, β, γ, δ and ε. Most normal human Hb contains two α-chains with two other β, γ, δ and ε sub-units. Depending on this, they can be classified as follows:

In the Embryo

1. **Gower 1($\zeta_2\varepsilon_2$) and Gower 2 ($\varepsilon_2\gamma_2$):** This is present in foetus in the first three months of pregnancy. There are two α-chains and two ε-chains, i.e., $\alpha_2\varepsilon_2$ in Gower 2.

2. **Hb Portland:** In this case, ε-chains and γ-chains are produced in access and bind with each other to form $\varepsilon_2\gamma_2$. Such foetuses do survive but for some time and commonly die after birth. Such a condition is known as hydrops foetalis.

In Adults

1. **Hb-A1 ($\alpha_2\beta_2$):** It constitutes of about 90–95 per cent of normal haemoglobin in adults. Normal adult Hb consists of two α-chains and two β-chains and they are expressed as $\alpha_2\beta_2$ (see Figure 5.5).
2. **Hb-A2 ($\alpha_2\delta_2$):** δ-chain synthesis begins late in the third trimester and in adults, normal range is 1.5–3.5 per cent (see Figure 5.5).
3. **Hb-A3:** It is present mainly in the old red cells and it is an altered form of Hb A. It constitutes to about 3–10 per cent of the total adult type.
4. **Hb-A_{1c}(Glycosylated Hb):** It is a glycosylated adult Hb present in increased percentage in the patients of diabetes mellitus. In a normal person, it is about 3–5 per cent but it increases to about 6–15 per cent of total Hb in case of people suffering from diabetes mellitus.

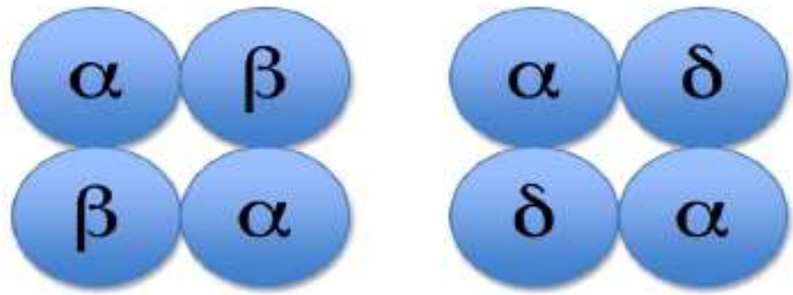


Fig. 5.5 *Structure of Hb in Adults*

Pathologic Mutant Variant

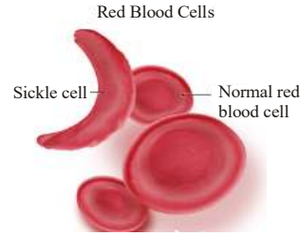
Hemoglobin variants are a part of the normal embryonic and foetal development, but may also be pathologic mutant forms of haemoglobin in a population, caused by variations in genetics. Some well-known haemoglobin variants, such as, sickle-cell anemia are responsible for diseases, and are considered haemoglobinopathies. Other variants cause no detectable pathology, and are thus, considered non-pathological variants.

- **Haemoglobin H (β_4):** It is a variant form of Hb, formed by a tetramer of β-chain, which causes a disease named 'thalassaemia major'. Beta thalassaemias are due to mutations in the HBB gene on chromosome 11, also inherited in an autosomal-recessive fashion. The severity of the disease depends on the nature of the mutation. Mutations are characterized as (β^o or β-thalassaemia major) if they prevent any formation of β-chains (which is the most severe form of β-thalassaemia) and they are characterized as (β^+ or β-thalassaemia intermedia) if they allow some β-chain formation to occur. In either case there is a relative excess of α-chains, but these do not

form tetramers, rather, they bind to the red blood cell membranes, producing membrane damage and at high concentrations they form toxic aggregates.

The a-thalassaemias involve the genes HBA1and HBA2, inherited in a Mendelian recessive fashion. It is also connected to the deletion of the 16p chromosome. Alpha thalassaemias result in decreased a-globin production, therefore fewer a-globin chains are produced, resulting in an excess of b-chains in adults and excess g-chains in newborns. The excess b-chains form unstable tetramers (called haemoglobin or Hb of 4 b- chains) which have abnormal oxygen dissociation curves as a- and b-chains are present in haemoglobin about 3 per cent of adult haemoglobin is made of a and b-chains. Just as with b-thalassaemia, mutations can occur which affect the ability of this gene to produce a-chains. This is known as d-thalassaemia.

- **Haemoglobin barts (γ_4):** It refers to the defect in α-chain gene. It causes less expression of α-chain which causes increased amount of γ-chain in respect to α-chain. This γ-chain forms tetramer. Haemoglobin S ($\alpha_2\beta_2^S$) is a case in which the Hb has hereditary defect in β-chain which results in defective Hb causing sickling of RBC. This condition is mostly known as sickle cell disease. This is caused in the homozygous condition (Refer Figure 5.6).



HBB Sequence in Normal Adult Hemoglobin (Hb A):

Nucleotide	CTG	ACT	CCT	GAG	GAG	AAG	TCT
Amino Acid	Leu	Thr	Pro	Glu	Glu	Lys	Ser
	3			6			9

HBB Sequence in Mutant Adult Hemoglobin (Hb S):

Nucleotide	CTG	ACT	CCT	GTG	GAG	AAG	TCT
Amino Acid	Leu	Thr	Pro	Val	Glu	Lys	Ser
	3			6			9

Fig. 5.6 *Sickle Cell*

- **Haemoglobin AS:** It refers to such a heterozygous condition in which one gene is normal adult Hb and another is sickle cell gene. An individual who has such a variety of cells suffers from sickle cell trait.

- **Haemoglobin C ($\alpha_2\beta_2^C$):** Some change in β-chain causes mild chronic haemolytic anaemia. The sixth position of β-chain which contains glutamic acid is replaced by lysine (Refer Figure 5.7).
- **Haemoglobin E ($\alpha_2\beta_2^E$):** If the twenty-sixth position of β-chain containing glutamic acid is replaced by lysine it causes mild chronic hemolytic anaemia.
- **Haemoglobin SC:** It is a heterozygous gene containing one form of sickle gene and another Hb C gene.
- **Haemoglobin D ($\alpha_2\beta_2^D$):** In this type of variant, the glutamic acid in the twenty-sixth position of β-chain is replaced glutamine.
- **Chronic Heinz Body Anaemia (CHBA)**: They are the unstable Hb variants which are formed due to amino acid substitution in the globin chain. The example of α-chain unstable variants are HbTorino, Hb Boyte Heights and Hb Point Philip. The examples of β-chain unstable variants are Hb Belfast, Hb St Luais and Hb Zurich. The γ-chain unstable variants include HbF Poole. All these type of variants cause haemolytic anaemia.
- **Variant with Increased Oxygen Affinity:** In this case, Hb binds very tightly with oxygen, so that oxygen cannot be removed easily. Examples of such variants are: α-chain variant (Hb Chesapeake) and β-chain variant (Hb Olympia).
- **Variant with Decreased Oxygen Affinity:** Hb has less tendency to bind with oxygen. For example, Hb Kansas, Hb Rothschild, Hb Hope.

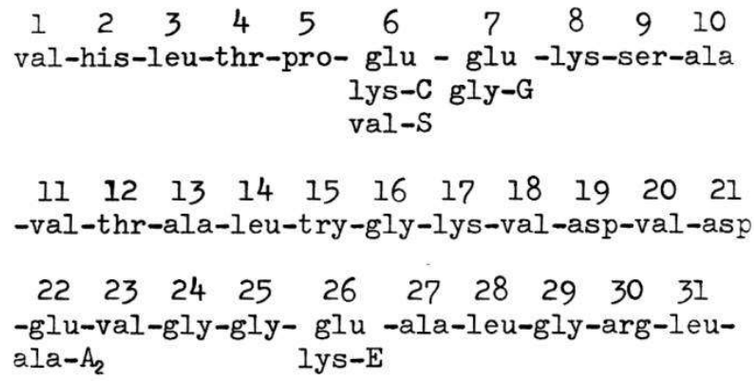


Fig. 5.7 *Mutant-Vaciants of Haemoglobin*

Derivatives of Haemoglobin

The nearest formal oxidation state of iron in Hb-O_2 is the +3 state, with oxygen in the–1 state (as superoxide .O_2^-). The diamagnetism in this configuration arises from the single unpaired electron on superoxide aligning anti-ferromagnetically from the single unpaired electron on iron, to give no net spin to the entire configuration, in accordance with diamagnetic oxy-haemoglobin from experiment.

1. **Carboxy Haemoglobin:** It is formed when carbon monoxide comes in contact with haemoglobin. The tendency of CO binding to Hb is 200 times more than O_2. This is harmful and life threatening.

2. **Methemoglobin:** In this case, iron changes to ferric state and is incapable of binding with oxygen. Normally, the Meth haemoglobin present in blood is about 1 per cent. The oxidation of Hb is stopped by the orientation of haeme pocket. The meth haemoglobin formed is reverted back to ferrous state by enzyme system present in the cell.
3. **Sulfhemoglobin:** It is formed when sulphur combines with the haeme of Hb. It is green in colour and is incapable of carrying oxygen.

Synthesis of Haemoglobin

Hb is synthesized in a complex series of steps. Formation of haeme part occurs in the mitochondria and cytosol of immature RBCs, while the globin part is synthesized by ribosome in cytoplasm. Production of Hb occurs during erythropoiesis of immature RBCs in red bone marrow. During erythropoiesis, even after loss of nucleus, the production of Hb continues till reticulocyte stage.

Synthesis of Haeme

Haemoglobin (Hb) is synthesized in a complex series of steps. The haeme part is synthesized in a series of steps in the mitochondria and the cytosol of immature red blood cells, while the globin protein parts are synthesized by ribosomes in the cytosol.

Following are the stages involved in synthesis of haeme (Refer Figure 5.8).

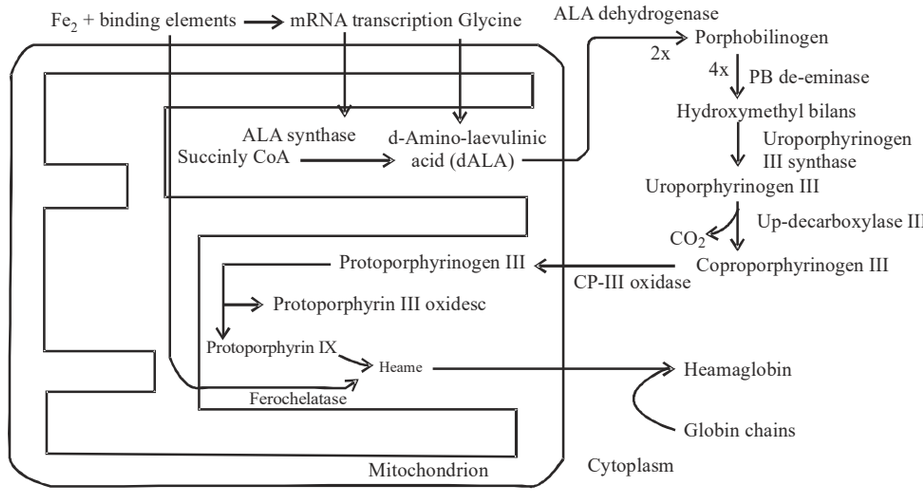


Fig. 5.8 *Synthesis of Haeme*

Biosynthesis of Porphyrin: Stages involved in the formation of porphyrin are:

Stage I

- **Synthesis of d-Amino Laevulinic Acid (δ-ALA):** It occurs in mitochondria. First, the succinyl CoA and glycine condense together to form α-amino-β-ketoadipic acid. After that on release of carbon-di-oxide, it forms δ-amino laevulinic acid. This reaction occurs in the presence of δ-ALA synthase. The coenzymes involved in this reaction are pyridoxal phosphate, pathothenic acid (forming CoA–SH) and Mg^{++}.

Stage II

- Synthesis of co-prophyrinogen III and I (cytosolic)
  - o Formation of porphobilinogen: d-ALA then moves into the cytoplasm from mitochondria. Two molecules of d-ALA undergo condensation reaction to form porphobilinogen in presence of enzyme d-ALA dehydrogenase. Co-factor involved is copper.
- Formation of uroporphyrinogen I and III
  - o Uroporphyrinogen I (minor series): In the presence of enzyme, porphobilinogen de-eminase (uroporphylinogen I synthase), four molecules of porphobilinogen condenses, losing four molecules of ammonia and form uroporphyrinogen I or hydroxymethyl bilane.
  - o Uroporphyrinogen III formation (major series): The cyclization of the uroporphyrinogen I occurs in the presence of uroporphyrinogen III synthase with deaminase. The product formed is called uroporphyrinogen III.
  - o Formation of co-prophyrinogen III: In presence of uroporphyrinogen III decarboxylase (removing carbon-di-oxide), the uroporphyrinogen III is decarboxylated to form coporphyrinogen III.

Stage III

- Formation of protoporphyrin IX

  After the formation of co-proporphyrinogen III, it enters mitochondria. In mitochondria, Coproporphyrinogen III is converted to protoporphyrinogen in the presence of coproporphyrinogen III oxidase. Protoporphyrinogen is then converted to protoporphyrin IX in presence of enzyme protoporphyrin III oxidase.

Stage IV

Fe^{2+} is inserted in central position of protoporphyrin IX by haeme synthetase to form haeme. Haeme is then coupled with protein globin to form haemoglobin.

5.2.2 Chlorophyll

Chlorophyll is any of several related green pigments found in the mesosomes of cyanobacteria and in the chloroplasts of algae and plants. Chlorophylls absorb light most strongly in the blue portion of the electromagnetic spectrum as well as the red portion. Conversely, it is a poor absorber of green and near-green portions of the spectrum. Hence chlorophyll-containing tissues appear green because green light, diffusively reflected by structures like cell walls, is less absorbed. Two types of chlorophyll exist in the photosystems of green plants: chlorophyll 'a' and 'b'.

Chlorophyll is the molecule that traps sunlight and is called a ***photoreceptor***. It is found in the chloroplasts of green plants and gives green colour to them. The basic structure of a chlorophyll molecule is a porphyrin ring, co-ordinated to a central atom. This is very similar in structure to the heme group found in hemoglobin, except that in heme the central atom is iron, whereas in chlorophyll it is magnesium. Like heme groups, chlorophylls are porphyrins found in plants. As such, they are tetrapyrroles that contain a metal ion at their core. Biosynthesis of the pigment starts

with the two important intermediates (uroporphyrinogen III and protoporphyrin IX) followed by the insertion of magnesium ion at its core. However, further modifications result in a variation of the different forms and specialization of the pigment in different organisms. Chemically, chlorophyll is composed of the following components: A nucleus of porphyrin (tetrapyrrole) that contains a chelated magnesium atom - The porphyrin head of the structure is composed of four rings of pyrrole with nitrogen arranged in a ring. Hydrocarbon chains linked through a group of carboxylic acid - The long hydrocarbon chain makes up the tail part of the structure. Chlorophyll pigments are bound by proteins that hold them in the right position. This positions them in the right alignment to trap light energy and transfer it during photosynthesis.

Chlorophylls are numerous in types, but all are defined by the presence of a fifth ring beyond the four pyrrole-like rings. Most chlorophylls are classified as chlorins, which are reduced relatives of porphyrins. They share a common biosynthetic pathway with porphyrins, including the precursor uroporphyrinogen III. Chlorophylls central magnesium atom coordinates with chlorin, a partially reduced porphyrin. The chlorin ring can have various side chains, usually including a long phytol chain. The most widely distributed form in terrestrial plants is chlorophyll 'a' and 'b'.

Molecular Structure of Chlorophyll

The molecular structure of chlorophyll a consists of a chlorin ring, whose four nitrogen atoms surround a central magnesium atom, and has several other attached side chains and a hydrocarbon tail.

Chlorophyll 'a' contains a magnesium ion encased in a large ring structure known as a chlorin. The chlorin ring is a heterocyclic compound derived from ***pyrrole***. Four nitrogen atoms from the chlorin surround and bind the magnesium atom. The magnesium center uniquely defines the structure as a chlorophyll molecule. The porphyrin ring of bacteriochlorophyll is saturated, and lacking alternation of double and single bonds causing variation in absorption of light. Side chains are attached to the chlorin ring of the various chlorophyll molecules. Different side chains characterize each type of chlorophyll molecule, and alters the absorption spectrum of light. The only difference between chlorophyll 'a' and chlorophyll 'b' is that chlorophyll 'b' has an aldehyde instead of a methyl group at the C-7 position. Chlorophyll 'a' has a long hydrophobic tail, which anchors the molecule to other hydrophobic proteins in the thylakoid membrane of the chloroplast. Structure of chlorophyll 'a' and 'b' are given below Figure 5.9.

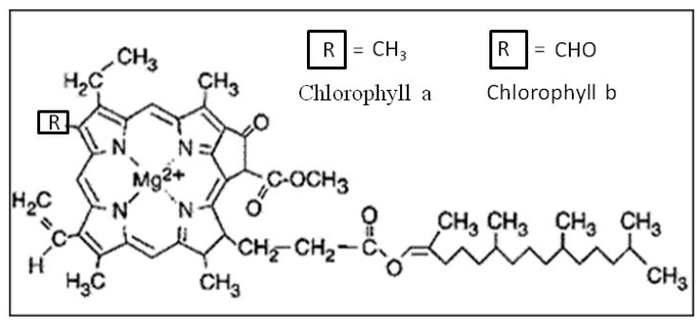


Fig. 5.9 *Structure of Chlorophyll 'a' and 'b'*

The structures of various chlorophylls are summarized in Table 5.1:

Table 5.1: *Structures of Different Types of Chlorophylls*

	Chlorophyll *a*	Chlorophyll *b*	Chlorophyll c_1	Chlorophyll c_2	Chlorophyll *d*	Chlorophyll *f*[14]
Molecular formula	$C_{55}H_{72}O_5N_4Mg$	$C_{55}H_{70}O_6N_4Mg$	$C_{35}H_{30}O_5N_4Mg$	$C_{35}H_{28}O_5N_4Mg$	$C_{54}H_{70}O_6N_4Mg$	$C_{55}H_{70}O_6N_4Mg$
C2 group	$-CH_3$	$-CH_3$	$-CH_3$	$-CH_3$	$-CH_3$	$-CHO$
C3 group	$-CH=CH_2$	$-CH=CH_2$	$-CH=CH_2$	$-CH=CH_2$	$-CHO$	$-CH=CH_2$
C7 group	$-CH_3$	$-CHO$	$-CH_3$	$-CH_3$	$-CH_3$	$-CH_3$
C8 group	$-CH_2CH_3$	$-CH_2CH_3$	$-CH_2CH_3$	$-CH=CH_2$	$-CH_2CH_3$	$-CH_2CH_3$
C17 group	$-CH_2CH_2COO-$Phytyl	$-CH_2CH_2COO-$Phytyl	$-CH=CHCOOH$	$-CH=CHCOOH$	$-CH_2CH_2COO-$Phytyl	$-CH_2CH_2COO-$Phytyl
C17–C18 bond	Single (chlorin)	Single (chlorin)	Double (porphyrin)	Double (porphyrin)	Single (chlorin)	Single (chlorin)
Occurrence	Universal	Mostly plants	Various algae	Various algae	Cyanobacteria	Cyanobacteria

Chlorophyll Synthesis

There are actually 2 main types of chlorophyll, named 'a' and 'b'. They differ only slightly, in the composition of a side chain (in 'a' it is -CH_3, in 'b' it is CHO). Both of these two chlorophylls are very effective photoreceptors because they contain a network of alternating single and double bonds, and the orbitals can delocalise stabilising the structure. Such delocalised polyenes have very strong absorption bands in the visible regions of the spectrum, allowing the plant to absorb the energy from sunlight.

In some plants, chlorophyll is derived from glutamate and is synthesised along a branched biosynthetic pathway that is shared with heme and siroheme. Chlorophyll synthase is the enzyme that completes the biosynthesis of chlorophyll 'a' by catalysing the reaction.

chlorophyllide 'a' + phytyl diphosphate $\rightleftarrows$ chlorophyll 'a' + diphosphate

This forms an ester of the carboxylic acid group in chlorophyllide a with the 20-carbon diterpene alcohol phytol. Chlorophyll 'b' is made by the same enzyme acting on chlorophyllide 'b'.

In Angiosperm plants, the later steps in the biosynthetic pathway are light-dependent and these plants are pale if grown in darkness. Non-vascular plants and green algae have an additional light-independent enzyme and grow green even in darkness. Chlorophyll itself is bound to proteins and can transfer the absorbed energy in the required direction. Protochlorophyllide, one of the biosynthetic intermediates, occurs mostly in the free form and, under light conditions, acts as a photosensitizer, forming highly toxic free radicals. Hence, plants need an efficient mechanism of regulating the amount of this chlorophyll precursor. In angiosperms, this is done at the step of AminoLevulinic Acid (ALA), one of the intermediate compounds in the biosynthesis pathway. Plants that are fed by ALA accumulate high and toxic levels of protochlorophyllide; so do the mutants with a damaged regulatory system.

Check Your Progress

1. Is porphyrin in haemoglobin?
2. What is the role of porphyrin in chlorophyll?
3. Define the haemoglobin.
4. What is photoreceptor and why it is called so?

5.3 PROSTAGLANDINS

Occurrence: These are a unique pharmacologically active lipids. These lipids are commonly found in tissues of mammals and also found in body fluids. These are also known as '***Eicosanoids***'.

Prostaglandins were discovered in human semen in 1935 by Swedish physiologist Ulf Von Euler. They give this name as they were recreated by the 'Prostate Glands'.

These were discovered by Swedish biochemists Sune K. Bergstron Bergt Ingemar Samuelsson and British biochemist Sir John Robert Vane, these three won Nobel Prize for physiology or medicine in 1982 for the isolation and identification of ***prostaglandins***.

5.3.1 Nomenclature, Classification, Biogenesis and Physiological Effectsof Prostaglandins

Nomenclature: Prostaglandins are derived from essential fatty acids and constitute a unique class of polyunsaturated, hydroxylated 20-carbon fatty acids categorized as ***eicosan***.

In the approved nomenclature, each prostaglandins name started with 'PG' followed by letter A to K, depending upon the nature and position of the substituents on the ring. Thus PGA to PGE and PGJ have a keto group in various positions on the ring and these are further distinguished by the presence or absence of double bonds or hydroxyl groups in various position on the group. PGE has two hydroxyl groups while PGK has two keto substitute on the ring. PGG and PGH are ***bicyclic endoperoxides***. An oxygen bridge between carbon 6 and carbon 9 distinguishes.

Prostacyclin (PGF): ThromboXane A (TXA) contains an unstable bicyclic oxygenated ring structure, while thromboXane B (TXB) has a stable **oxane ring**. In addition, all prostaglandins have a hydroxyl group in the S-configuration on carbon 15 and 9 trans-double bond at carbon 13 of the alkyl substituent (R_2).

Further, a numerical subscript (1 to 8) is used to denote the total number of double bond in the alkyl substituents. A Greek subscript (α or β) is used with prostaglandins of the PGF series to describe the stereochemistry of the hydroxyl group on carbon 9.

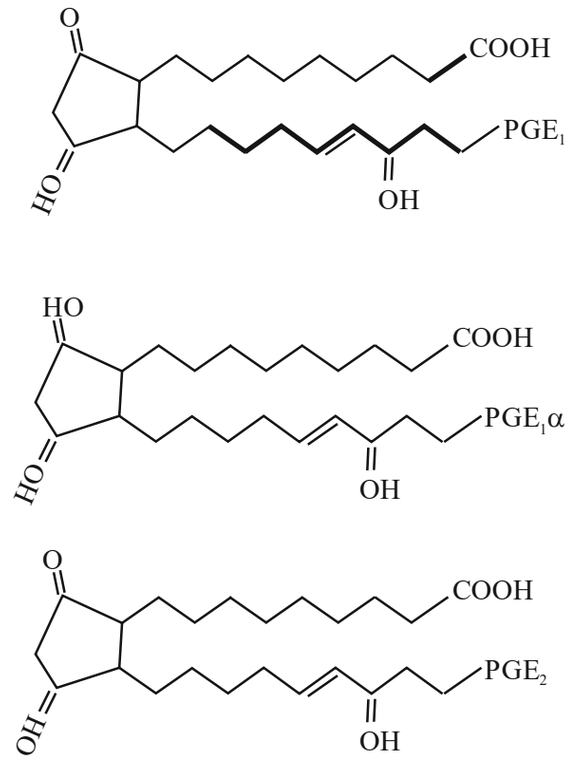


Fig. 5.10 *Structure of PGE_1, $PGE_1\alpha$ and PGE_2*

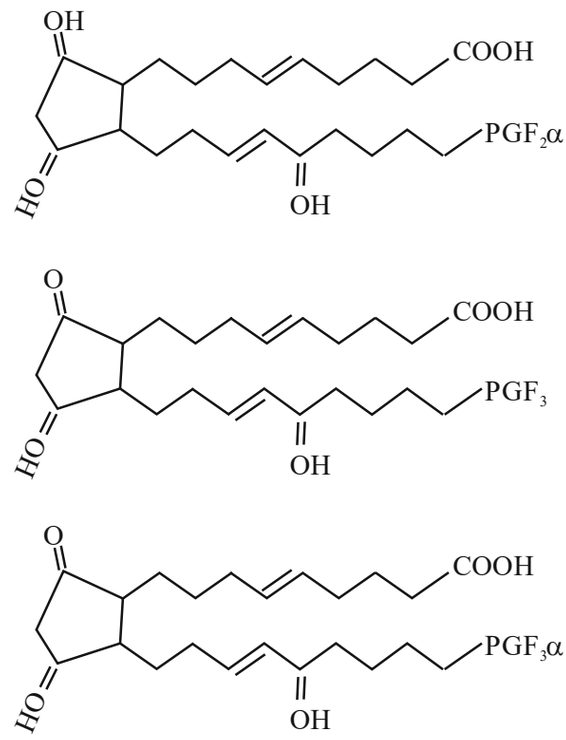


Fig. 5.11 *Structure of PGF_2, PGE_3 and $PGF_{3\alpha}$*

The number of (=) double bonds mainly depends on fatty acid.

Classification of Prostaglandins: Prostaglandins are classified as PGE, PGF, PGD, PGI.

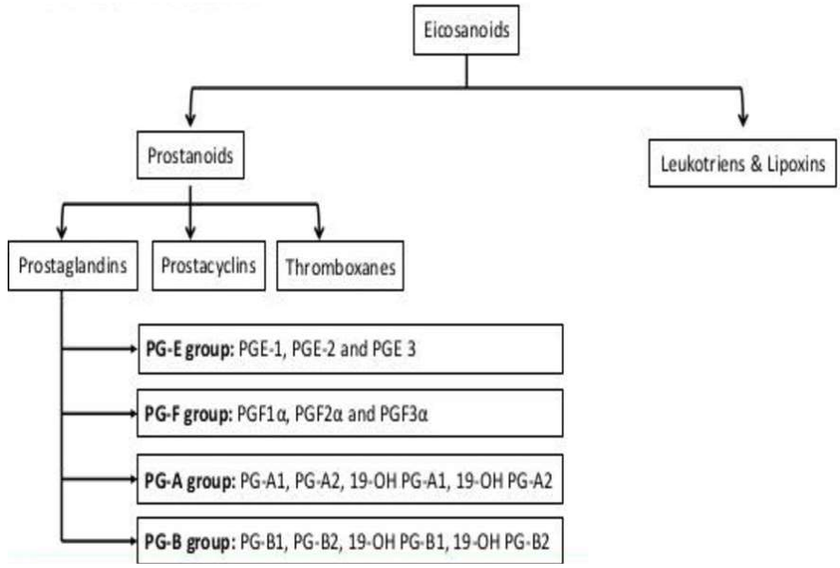


Fig. 5.12 *Classification of Prostaglandins*

Biogenesis and Physiological Effects

During cell activation, a large spectrum of lipid mediators is product through membrane phospholipid breakdown by the action of diverse ***phospholipases.***

- Arachidonic acid (5, 8, 11, 14- eicosatetraenoic acid) is the precursor for most of the prostaglandins in human.
- It occurs in the endoplasmic reticulum in the following stages.
  1. **Prospholipase A_2** release arachidonic acid from membrane bound phospholipids. Some hormones epinephrine or bradykinin stimulates this reaction.
  2. Oxidation and cyclization of arachidonic acid to PGG_2 by cyclooxygenase 1 & 2.
  3. PGG_2 is then converted to PGH_2 by a reduction in presence of glutathione dependent peroxidase.
  4. PGH_2 (Refer Figure 5.13) serves as immediate precursor for the synthesis of different types of prostaglandins.

NOTES

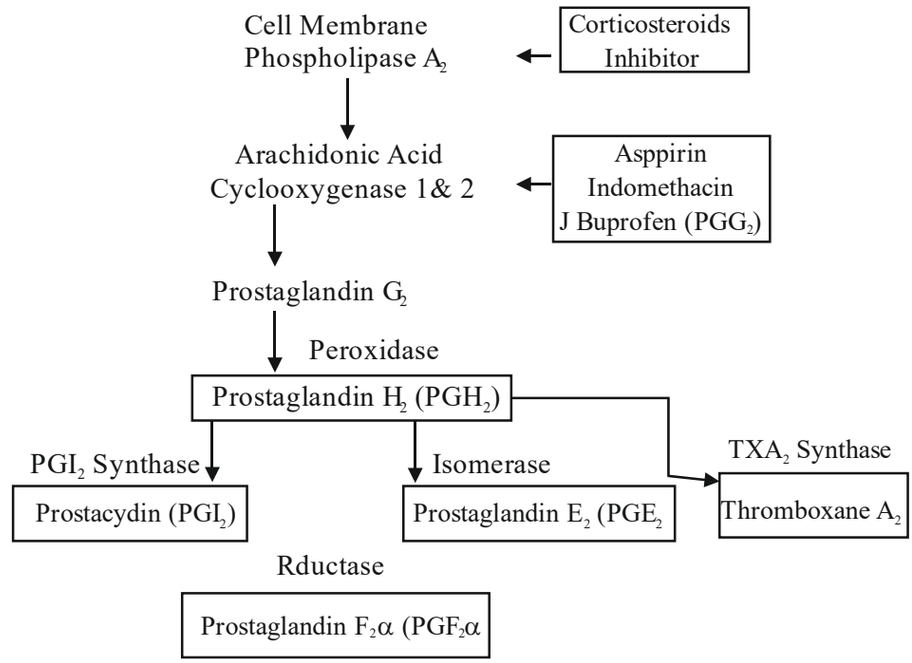


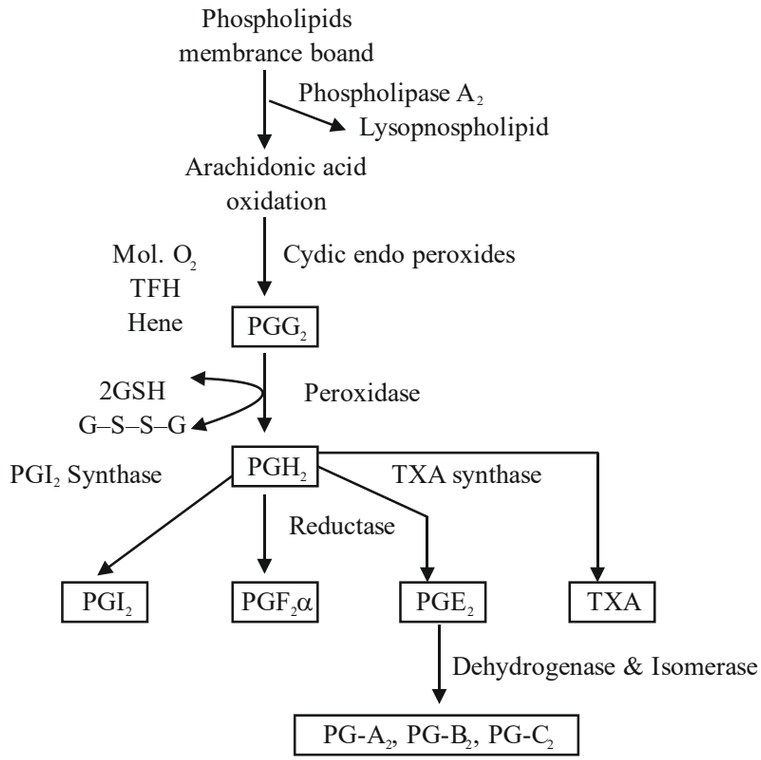
Fig. 5.13 *Formation of PGI_2, PGE_2 and $PGF_{2\alpha}$*

Physiological Effects

1. **Regulation of Blood Pressure:** PGE_2 and PGI_2 are vasodilators in vascular bed which results in increased blood flow and decrease peripheral resistance which leads to lower blood pressure.
2. **Inflammation:** PGE_1 and PGE_2 increase the symptoms of inflammation (redness, swelling etc.) due to vasodilation.
3. **Reproduction:** PGE_2 and $PGF_{2\alpha}$ causes contraction of uterine smooth muscle in pregnant woman.
4. **Pain and Fever:** Fever producing agents known as pyrogens, promotes prostaglandins biosynthesis which leads to formation of PGE_2 in hypothalamus and causes fever associated with pain.
5. **Regulation of Gastric Secretion:** Prostaglandin stimulates pancreatic reaction and increases the mortality of intestine and cause diarrhoea. It also inhibits gastric secretion.
6. **Influence on Immune System:** PGE decreases immunological function of B and T lymphocytes.
7. **Effect on Respiratory Function:**
   - PGEs causes bronchial smooth muscle relaxation
   - PGFs causes bronchial smooth muscle constriction, thus, PGE and PGF oppose the actions of each other in the lung.
8. **Influence or Renal Function:** It increases GFR and promotes urine output.
9. **Effect on Platelet Aggregation:** PGI2 inhibits platelet aggregation. TXA2 and PGE2 promotes platelet aggregation and blood clotting, that leads to thrombosis.

Synthesis of PGE2 and PGF2α

- Arachidonic acid (5, 8, 11, 14 eicosatetraenoic acid) is the precursor for most of the prostaglandins in humans.
- It occurs in the endoplasnic reticulum.



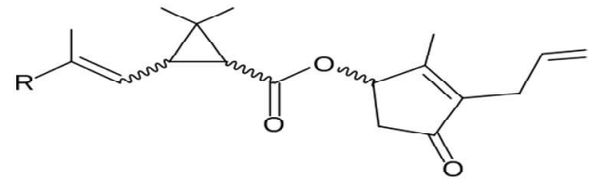
Check Your Progress

5. What do you understand by eicosanoids?
6. When ThromboXane A (TXA) is stable?
7. Give the name of fever producing agents.

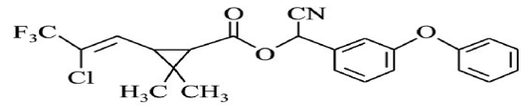
5.4 SYNTHESIS AND REACTIONS OF PYRETHROIDS AND ROTENONE

Synthesis of Pyrethroids: Pyrethroids are synthetic insecticides analogs of pyrethrins, these are used to control insect, pests, etc., in agriculture and public health .They are derived from naturally occurring Pyrethrins and consist of two basic structures, an acid and an alcohol. When high doses of pyrethroids are ingested orally, central nervous system symptoms may occur. Pyrethroids produces acute toxicity.

NOTES

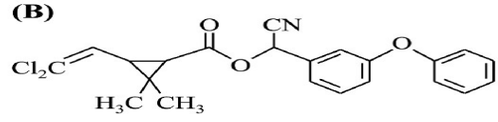


(A)



Lambda-cyhalothrin

(B)



Cypermethrin

Type I – Shorter duration of effects cause severe fine tremor.

Type II – Longer acting enhanced by addition of cyano group causes coarse tremor.

Type I – Pesticides – Pyrethrins, Bioallethrin, Cismethrin.

Type II – (Cyano Pesticides)- Fenvalerate, Cyhalothrin, Deltamethrin, Cypermethrin.

Pyrethrins

Pyrethrum is derived from the dried flowers of the plant Chrysantheum cineariaefolium. The active insecticidal component of the dried flowers is known as Pyrethrins, Chemically, pyrethrins are organic esters formed by the combination of two carboxylic acids and three keto alcohols (Refer Figure 5.16).

The properties of Pyrethrins are:

- Rapid Action
- Low Mammalian Toxicity
- Broad Spectrum Activity
- Lack of Persistence
- Repellency

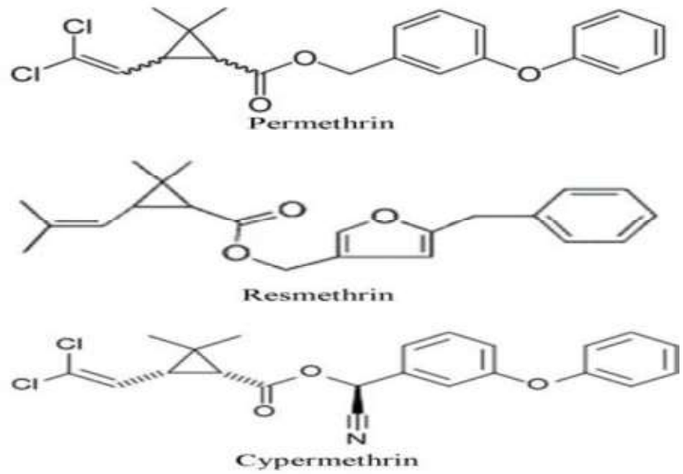


Fig. 5.14 *Structures of Permethrin, Resmethrin and Cypermethrin*

Allethrin is prepared by its esterification of synthetic Chrysanthemic acid with the alcohol allethrolone. Removal of keto group from Allethrin gave another synthetic pyrethroid, known as 'Bioallethrin'.

Bioresmethrin is an extremely active insecticide. This is photosensitive and was not persistent. When the iso butenyl group of bioresmethrin was replaced by the dichlorovinyl group, the resultant compound is NRDC134 which was more toxic to house flies.

Permethrin was active against houseflies and mustard beetles and showed much greater photostability and a moderately persistent insecticide. It was the first synthetic pyrethroid effective as a seed treatment against wheat bulb fly.

The corresponding chloro derivative is known as cypermethrin which is a broad spectrum insecticide with good residual activity on plants.

A survey of the esters of furyl methanol led to the discovery of insecticidal activity in a group of phenyl acetic acid esters known as fenvalerate. Fenvalerate is a mixture of 4 isomers and is used against a wide range of pests and relatively stable in light. Another phenyl acetic acid esters flucythrinate and fluvalinate**.**

Bromination of the double bonds on decamethrin and cypermethrin gave tralomethrin and tralocythrin.

'Lambda Cyhalothrin' has a comparatively high mammalian toxicity. It is effective at very low doses against major insect pests in many crops. Little hazard to honey bees and this represents an important advantage over insecticides which are highly toxic to honey bees. At normal rates cyhalothrin shows low toxicity to birds with no accumulation in eggs or tissues and no effect on earthworms.

Tefluthrin is the first pyrethroid effective as a soil insecticide at doses of 12-150 g. It is formulated as granules and may also be applied as foliar spray or seed dressing.

Reaction of Pyrethroids

1. The symptoms of insects poisoned by pyrethroids clearly show that the chemical attacks on the insect's nervous systems.
2. Pyrethroids cause hyper excitation followed by convulsions and death in arthropods.
3. The rate and mechanism of metabolism has a major influence on the toxicology of a compound.
4. In (rats) mammals, pyrethroids are very rapidly metabolized by ester cleavage, oxidation hydroxylation.
5. The synthetic pyrethriods are very expensive to prepare on a tonnage basis.
6. The high insecticidal activity and low mammalian toxicity of pyrethroids are especially significant now that compounds stable to light and oxygen are potentially available.
7. Their toxicity to fish is high. They are rapidly degraded in soil and have no detectable ill effects on soil microflora and microfauna. They are not active against mites.
8. The major symptoms of pyrethroid poisoning in insects may be accounted by effects on the kinetics of nerve membrane sodium channels.
9. The mean open times of these channels are prolonged with consequent hyperactivity of nerves.
10. The synthetic pyrethroids have been found to be useful as early season sprays to control the variety of insects that occur on cotton including boll worms, leaf worms, jassids, thrips and whitefly.

Rotenone

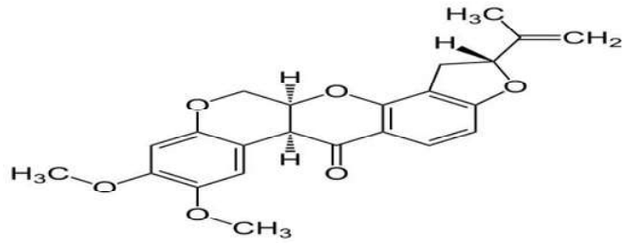
Preparation and Reactions

Molecular Formula $C_{23}H_{22}O_6$

Rotenone is a natural active ingredient with broad insecticidal and a few acaricidal properties, derived from the roots or rhizomes of several tropical plants such as, *Derris*, *Amorpha*, *Lonchocarpus* and *Tephrosia*. The root and stem part of *D. scandens* were reported as showing good antibacterial, antifungal and antialgal properties.

Rotenone is colourless to brownish or a white to brownish-white crystalline solid and odourless naturally found organic compound isoflavone. This is used as broad spectrum insecticide, piscicide and pesticide. Naturally it is found in seed roots and stem of many plants such as Derris, Amorpha, Lonchocarpus and Tephorsia belongs to Fabaceae family. Rotenoids are the group of compounds which has one of compound is Rotenone.

Rotenone consists of 1,2,12,12a-tetrahydrochromeno[3,4-b] furo[2,3-h] chromen-6(6aH)- one substituted at position 2 by a prop-1-en-2-yl group and at positions 8 and 9 by methoxy groups (the 2R,6aS,12aS-isomer).



The synthesis of rotenone involves two key transformations, the first of which is a Pd π-allyl mediated cyclisation for the construction of the dihydrobenzofuran skeleton. The second is a 6-*endo*- hydroarylation which yields the chromene as a precursor to rotenone. The synthesis of rotenone was achieved in 17 steps from resorcinol. The active chemical component was first isolated by a French botanist Emmanuel Geoffroy in 1895 and called it nicouline, A Japanese chemist Nagai isolated a pure crystalline compound from Derris elliptica which he called rotenone.

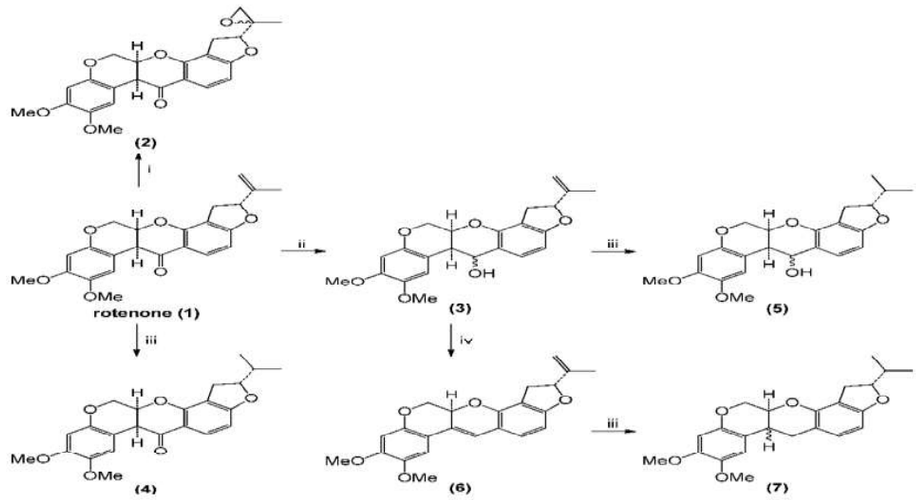


Fig. 5.15 *Synthesis of Rotenone*

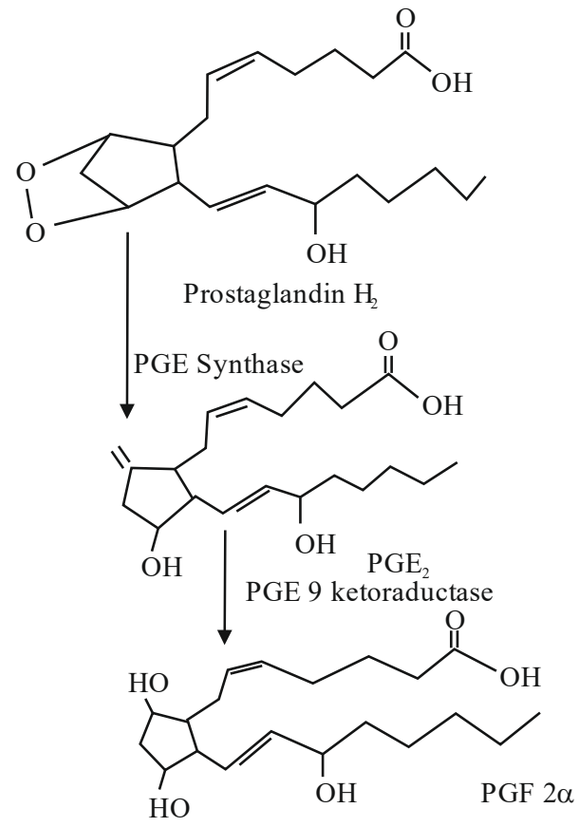
Reactions

- Rotenon works by interfering with the electron transport chain in mitochondria.
- It inhibit the transfer of electrons from iron sulfur centres in complex I to Ubiquinone. This interferes with NADH during the creation of usable cellular energy (ATP).
- Cellular oxygen is reduced to the radical, creating a reactive oxygen species, which can damage DNA and other components of the mitochondria. It has three chiral centres thus has complex stereochemistry.
- Rotanone is applied directly to water to manage fish population in lakes, ponds, reservoirs, rivers, streams, and in aquaculture. The chemical can be applied to an entire water body to achieve a complete kill or a protection of a water body to achieve a partial kill.
- Rotenone is used to catch fish. Small-scale sampling with rotenone is used by fish researchers studying the biodiversity of marine fishes.

- Rotenone also used to treat head lice on humans, and parasitic mites on chickens.

Reaction of Prostaglandins Synthesis

Reaction



Check Your Progress

8. What is pyrethrins?
9. Give the properties of pyrethrins.
10. Define the tefluthrin.
11. Give the molecular formula of rotenone.

5.5 ANSWERS TO 'CHECK YOUR PROGRESS'

1. The porphyrins are heterocyclic ring structures that include four pyrrole rings joined together through carbon (methenyl) bridges. The most abundant porphyrins in nature are found in hemoglobin and the chlorophylls. In the centre of porphyrins a metal atom is chelated to the nitrogen atoms of the pyrrole units.
2. Chlorophyll is the green pigment in plants, algae, and cyanobacteria, i.e., essential for photosynthesis. Its central structure is an aromatic porphyrin

or chlorine (reduced porphyrin) ring system with a sequestered magnesium atom. A fifth ring is fused to the porphyrin.

3. Haemoglobin is a conjugated chromoprotein which contains haeme as prosthetic group and globin as the apoprotein part. Conjugated proteins are proteins which have a protein part known as apoprotein and a non-protein part known as prosthetic group. Chromoproteins are proteins which are coloured.
4. Chlorophyll is the molecule that traps sunlight and is called a photoreceptor. It is found in the chloroplasts of green plants and gives green colour to them. The basic structure of a chlorophyll molecule is a porphyrin ring, co-ordinated to a central atom.
5. These are a unique pharmacologically active lipids. These lipids are commonly found in tissues of mammals and also found in body fluids. These are also known as 'Eicosanoids'.
6. ThromboXane A (TXA) contains an unstable bicyclic oxygenated ring structure, while thromboXane B (TXB) has a stable oxane ring.
7. Fever producing agents known as pyrogens, promotes prostaglandins biosynthesis which leads to formation of PGE_2 in hypothalamus and causes fever associated with pain.
8. Pyrethrum is derived from the dried flowers of the plant Chrysantheum cinearіaefolium. The active insecticidal component of the dried flowers is known as Pyrethrins, Chemically, pyrethrins are organic esters formed by the combination of two carboxylic acids and three keto alcohols.
9. The properties of Pyrethrins are:
   - Rapid Action
   - Low Mammalian Toxicity
   - Broad Spectrum Activity
   - Lack of Persistence
   - Repellency
10. Tefluthrin is the first pyrethroid effective as a soil insecticide at doses of 12-150 g. It is formulated as granules and may also be applied as foliar spray or seed dressing.
11. Rotenone molecular formula is $C_{23}H_{22}O_6$.

5.6 SUMMARY

- Metal complexes derived from porphyrins occur naturally. One of the best-known families of porphyrin complexes is heme, the pigment in red blood cells, a cofactor of the protein haemoglobin.
- A porphyrin without a metal-ion in its cavity is a free base. Some iron-containing porphyrins are called hemes. Heme-containing proteins, or haemoproteins, are found extensively in nature.

- The organic part is a porphyrin ring based on porphin (a tetrapyrrole ring), and is the basis of a number of other important biological molecules, such as, chlorophyll and cytochrome.
- Oxygenated haemoglobin (found in blood from arteries) is bright red, but without oxygen present (as in blood from veins), haemoglobin turns a darker red.
- The porphyrins are heterocyclic ring structures that include four pyrrole rings joined together through carbon (methenyl) bridges. The most abundant porphyrins in nature are found in haemoglobin and the chlorophylls.
- As chlorophyll they are at the heart of photosynthesis. They are involved in the critical energy conversion step of photosynthesis.
- Haeme is iron-containing porphyrin nucleus. The porphyrins are complex in nature, having a 'Tetra-Pyrrole' (four pyrrole rings) structure which are linked by – CH = bridges called 'Methynyl' or 'Methylidine' bridges.
- Haemoglobin variants are a part of the normal embryonic and foetal development, but may also be pathologic mutant forms of haemoglobin in a population, caused by variations in genetics. Some well-known haemoglobin variants such as sickle-cell anemia are responsible for diseases, and are considered haemoglobinopathies.
- Haemoglobin SC is heterozygous gene containing one form of sickle gene and another Hb C gene.
- Hb is synthesized in a complex series of steps. Formation of haeme part occurs in the mitochondria and cytosol of immature RBCs, while the globin part is synthesized by ribosome in cytoplasm. Production of Hb occurs during erythropoiesis of immature RBCs in red bone marrow.
- Fe^{2+} is inserted in central position of protoporphyrin IX by haeme synthetase to form haeme. Haeme is then coupled with protein globin to form haemoglobin.
- Prostaglandins are derived from essential fatty acids and constitute a unique class of polyunsaturated, hydroxylated 20-carbon fatty acids categorized as eicosan
- The number of (=) double bonds mainly depends on fatty acid.
- Release arachidonic acid from membrane bound phospholipids. Some hormones epinephrine or bradykinin stimulates prospholipase A_2 reaction.
- PGE2 and PGI2 are vasodilators in vascular bed which results in increased blood flow and decrease peripheral resistance which leads to lower blood pressure.
- Prostaglandin stimulates pancreatic reaction and increases the mortality of intestine and cause diarrhoea. It also inhibits gastric secretion.
- Arachidonic acid (5, 8, 11, 14 eicosatetraenoic acid) is the precursor for most of the prostaglandins in humans.
- Pyrethroids are synthetic insecticides analogs of pyrethrins, these are used to control insect, pests, etc., in agriculture and public health .They are derived

from naturally occurring Pyrethrins and consist of two basic structures, an acid and an alcohol.

- Bioresmethrin is an extremely active insecticide. This is photosensitive and was not persistent. When the iso butenyl group of bioresmethrin was replaced by the dichlorovinyl group, the resultant compound is NRDC134 which was more toxic to house flies.
- Permethrin was active against houseflies and mustard beetles and showed much greater photostability and a moderately persistent insecticide. It was the first synthetic pyrethroid effective as a seed treatment against wheat bulb fly.
- The high insecticidal activity and low mammalian toxicity of pyrethroids are especially significant now that compounds stable to light and oxygen are potentially available.

5.7 KEY TERMS

- **Porphyrins:** Porphyrins are a group of heterocyclic, macrocycle organic compounds, composed of four modified pyrrole subunits which contain interconnected at their α-carbon atoms via 'Methine Bridges' (=CH").
- **Heme:** While the haemoglobin and myoglobin molecules are very large, complex proteins, the active site is actually a non-protein group called heme.
- **Chlorophyll:** Chlorophyll is the green pigment in plants, algae, and cyanobacteria, i.e., essential for photosynthesis.
- **Methylidine Bridges:** Haeme is iron-containing porphyrin nucleus. The porphyrins are complex in nature, having a 'Tetra-Pyrrole' (four pyrrole rings) structure which are linked by –CH = bridges called 'Methynyl' or 'Methylidine' bridges.
- **Bioallethrin:** Allethrin is prepared by its esterification of synthetic Chrysanthemic acid with the alcohol allethrolone. Removal of keto group from Allethrin gave another synthetic pyrethroid, known as 'Bioallethrin'.
- **Lambda Cyhalothrin:** 'Lambda Cyhalothrin' has a comparatively high mammalian toxicity. It is effective at very low doses against major insect pests in many crops.

5.8 SELF-ASSESSMENT QUESTIONS AND EXERCISES

Short-Answer Questions

1. Define the prophyrins.
2. What is the role of porphyrin in haemoglobin?
3. Give the specific definition of haemoglobin.
4. What is chlorophyll?

NOTES

5. Give the biogenesis and physiological effect of protaglands.
6. Define the term pyrethroids.
7. What do you understand by rotenones?

Long-Answer Questions

1. Elaborate on the porphyrins and what is the role of porphyrin in haemoglobin and chlorophyll.
2. Illustrate the structure and synthesis of haemoglobin and chlorophyll.
3. Describe the prostaglandins giving biogenesis and physiological effects.
4. Explain in detail about the PGE_2 and $PGF2_\alpha$ with the help of relevant reactions.
5. Analyse the synthesis and reactions of pyrethroids and rotenones.

5.9 FURTHER READING

Krishnaswamy, N. R. 1999. *Chemistry of Natural Products A Unified Approach*. Himayatnagar, Hyderabad: Universities Press.

Bhat, Sujata V., B.A. Nagasampagi, Meenakshi Sivakumar · 2005. *Chemistry of Natural Products*. Narosa Publishing House

Rahman,Atta-ur. 2020. *Studies in Natural Products Chemistry.* Volume 67. Amsterdam, Netherlands: Elsevier Science.

Cooper, Raymond. , George Nicola. 2014. *Natural Products Chemistry-Sources, Separations and Structures.* London UK: Taylor & Francis.